CHEMISTRY IN RELATION TO BIOLOGY & MEDICINE WITH ESPECIAL REFERENCE TO INSULIN & OTHER HORMONES

By JOHN JACOB ABEL

THE WILLARD GIBBS LECTURE

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Delivered before the Seventh Midwest Intersectional Meeting on the occasion of the award to Dr. Abel of the Willard Gibbs Gold Medal by the Chicago Section of the American Chemical Society, May 27, 1927, and here reprinted from *Science*, October 7 and 14, 1927 as a tribute to the distinguished author, and as a memorial of the early stimulus he gave to the activities of the Williams and Wilkins Company in science publishing.

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PUBLISHER'S PREFACE

THIS LIMITED EDITION of Dr. Abel's famous *Willard Gibbs Lecture* has been undertaken, like so many of Dr. Abel's own endeavors, as a labor of love. The undertaking was prompted by a desire to do honor to Dr. Abel's memory in an appropriate form. It was thought peculiarly fitting for these companies to pay such tribute, for Dr. Abel played a vital part in shaping their development. Aware that in honoring Dr. Abel we honor ourselves, we recite the facts that seem to justify us in doing so.

In the year 1909, Dr. Abel was desirous of launching *The Journal of Pharmacology and Experimental Therapeutics.* In casting about for a printer he encountered Edward B. Passano, president of what was then known as Williams & Wilkins Company, a pioneer in serving the scientific world as printer, but not as publisher. Dr. Abel laid before Mr. Passano the lack of American facilities for science publication, and showed to him the potentialities of this field of endeavor.

The immediate result was that the then printing company did undertake to publish Dr. Abel's journal and has continued to do so ever since. A further result was the inception, during the next decade, of a number of new American science journals. Book publication was added in 1920, and by January 1925 the publishing branch of the business had grown to a point where separate entity for it was imperative. The old name, with a slight change, was retained for the publishing business, and the printing branch took the style Waverly Press, Inc. Today each company is the *alter ego* of the other, and both are still under the presidency of Edward B. Passano.

Allusion to this bit of history is made only to emphasize Dr. Abel's contribution. Doubtless many others in the first decade of the century felt the lack of American facilities for publication, but he had the foresight to look into the future a little more clearly, and the lucid perception that enabled him to convey his vision to another and thus lay foundations for its practical fulfilment.

The establishment of a publishing house would probably not have been reckoned by Dr. Abel himself as among his achievements. Yet there can be no question that he planted the seed from which one germinated. If the fact adds nothing to his stature, it is because that stature is already great.

Grateful acknowledgment is made to *Science* magazine and to Dr. Abel's heirs for permission to make this edition and place it in the hands of persons deemed most likely to be appreciative.

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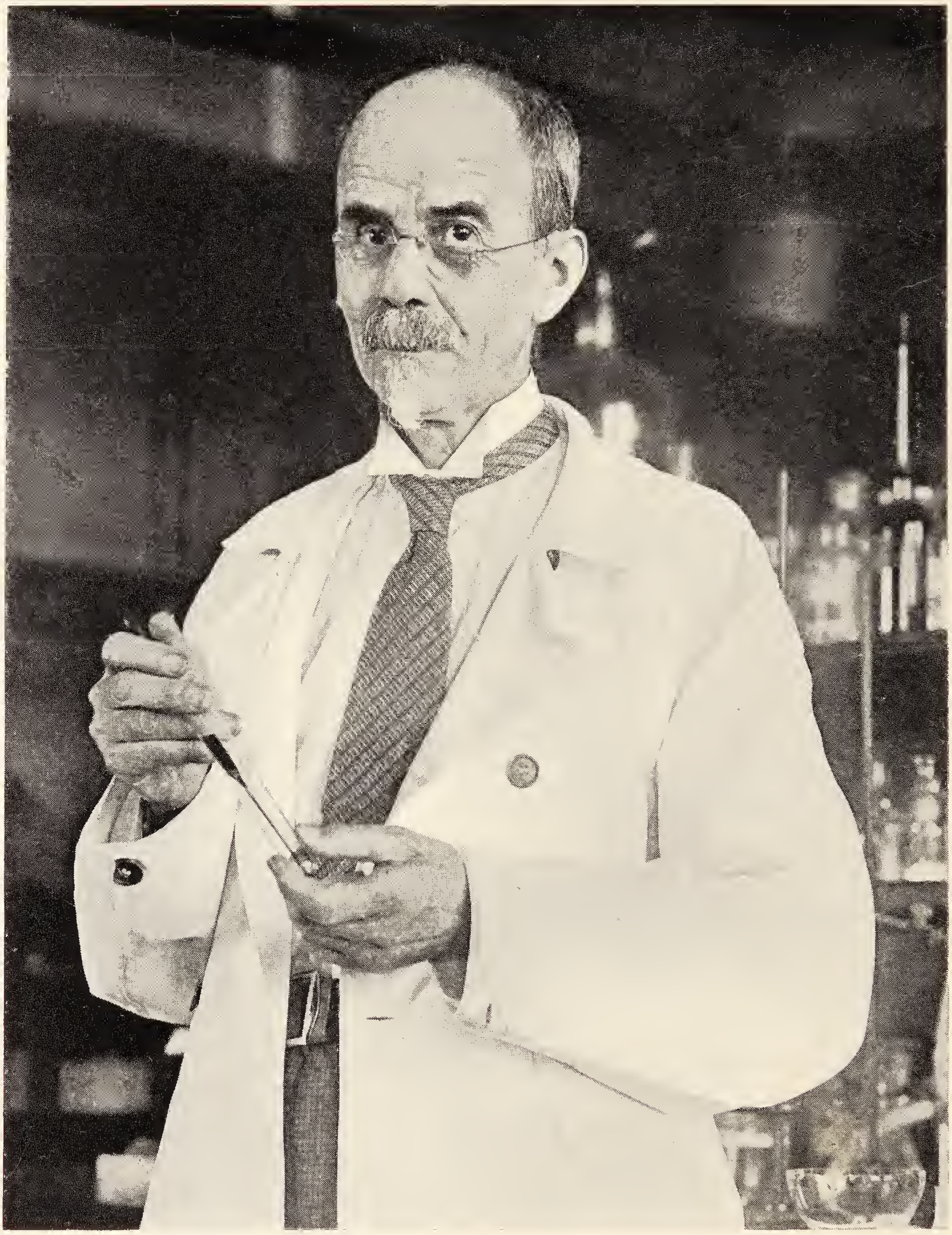


John J. Abel

JOHN JACOB ABEL

In Baltimore, on May 26, 1938, the long career of a great pioneer figure in American experimental medicine ended. Besides being distinguished for his many notable contributions in his chosen fields, John J. Abel will be remembered as the "Father of American Pharmacology" and the founder of a school of pharmacologists. Those with whom he came into intimate contact will never forget his magnetic yet simple personality, his unquenchable devotion to research, his high idealism, indomitable enthusiasm and optimism, his remarkable capacity for turning apparent defeat into victory and the unique ability he had developed for dealing with minute amounts of complex chemical substances of biological importance.

Abel was born on May 19, 1857, near Cleveland. His family came from the Rhine Valley of the Palatinate. He had no scientific forbears on either side. He received his Ph.B. degree from the University of Michigan in 1883, but had an interim of three years in his college course, during which he served as principal of a high school at LaPorte, Indiana, 1879–1880, where he taught Latin, mathematics, physics and chemistry. From 1880 to 1882 he was superintendent of the public schools at LaPorte. Abel must have looked back on these years with satisfaction, for it was in LaPorte that he met a teacher in his school, whom he once described as a "very sweet mild little lady with a great deal of force." This lady, Mary Hinman, became his wife on July 10, 1883, was his companion for fifty-five years, and made possible the life of intense devotion to research which he chose to live. All who knew Abel at all intimately realize the important rôle which Mrs. Abel played in his successful career.

It was characteristic of the man, that after choosing scientific medicine as a life work, he submitted himself to a prolonged, broad, fundamental training. After a year in graduate study with Newell Martin at the Hopkins, seven years were spent in some of the leading universities of Europe studying chemistry and medicine. His teachers during this period are remembered for their distinction in their various fields: in anatomy, His, Braume and Schwalbe; in physiology, Carl Ludwig; in pharmacology, Oswald Schmiedeberg; in biochemistry, Drechsel, Hoppe-Seyler and von Nencki; in chemistry, Wislicenus; in pathology, Arnold and v. Recklinghausen; and in clinical branches such men as Wagner, Kussmaul, Erb and Naunyn. His breadth of training is also evidenced by the many universities in which he studied: Leipzig, Strassburg, Heidelberg, Berne, Vienna, Würzburg, Berlin and Paris. In 1888, he received the M.D. degree from the University of Strassburg. Knowing that he had to earn his living and realizing the lack of full-time opportunities in scientific medicine in this country half a century ago, Abel now spent a year "walking the wards" in Vienna to prepare for the possibility of having to practice medicine as the only outlet of a scientific career. It must have been a great relief when he was asked to occupy the chair of materia medica and therapeutics in the University of Michigan, where he remained for two years. In 1893, he came to The Johns Hopkins Medical School as professor of pharmacology, a chair he occupied until his retirement in July, 1932. On his retirement as emeritus professor, he served as director of the Laboratory of Endocrine Research, a special creation for his work, and here he pursued active research until a few days before his death. It was typical of the man that as soon as he was established in this laboratory, a new problem, having nothing to do with endocrines, captured his attention.

During his first years as professor of pharmacology at the Hopkins, he taught physiological chemistry as well as pharmacology, and for many years thereafter the former subject remained under his direction until his former associate, Walter Jones, was made professor of physiological chemistry in 1908.

Abel's contributions to medical science cover a wide range of subjects and extend over a period of a half century. Space will not permit nor is this the place for any detailed analysis or evaluation of his work. The main theme which seems to have attracted his attention early and to which he returned again and again was the isolation of the active constituents of various endocrine glands. That he early appreciated the great significance of such contributions is best shown by a quotation from one of his addresses. "The actual findings of definite and specific chemical principles in the organs of internal secretion has in each case an importance in the way of explaining and correlating a large number of disconnected facts, only to be likened to the discovery of the etiological cause of an infectious disease." His interest in the internal secretions was intense and he was fond of emphasizing the importance of the active pharmacological substances present in our bodies in the aphorism: "We are walking drug stores."

In 1895, nothing was known about the chemical nature of any of the active principles of the glands of internal secretion. Abel started work on the extremely difficult problem of the isolation of the pressor constituent of the suprarenal medulla. The result of this work was the isolation of the hormone, not in the form of its free base but in that of a monobenzoyl derivative. As a result of this pioneer work of Abel, others later obtained the pure crystalline hormone, and this has been followed by thousands of papers by other investigators dealing with the various chemical, physiological and therapeutic aspects of epinephrine. Some years later, when examining a specimen of a tropical toad, which exudes from its skin glands a creamy secretion used as an arrow poison, Abel noticed that this secretion made on a scalpel a peculiar greenish blue discoloration. Remembering that he had seen this color years before on a scalpel used in cutting the medulla of the suprarenal gland, he set to work and soon isolated from this external secretion the now familiar epinephrine.

This pioneer work of Abel in the isolation of epinephrine, the first hormone to have its chemical nature elucidated, his brilliant success in the isolation of the pancreatic hormone, insulin, as a beautifully crystalline compound from inert constituents of pancreatic extracts, and his ingenious invention of the method of dialyzing the circulating blood of a living animal are achievements sufficient to class him as an investigator of first rank. Many other researches were conducted along these lines of the chemical nature of the principles of internal secretion, many of which he never considered worthy of record. His very few general addresses—the Mellon, Harvey, Kober and Willard Gibbs lectures—all deal with the internal secretions and emphasize the importance of chemistry in medical research; they are all examples of painstaking and prolonged preparation in presenting the subject in an erudite and scholarly manner.

In between the periods when intensive effort was directed to problems of isolation of pure crystalline active principles, we find researches of a totally different type—a rather unusual combination for a single individual. The behavior of frog's muscle to acids, pharmacological study of the phthaleins, the efficacy of new antimony compounds in experimental trypanosomiasis, convulsions in frogs produced by acid fuchsin, the influence of the lymph hearts upon the action of drugs in cardiectomized frogs, and the studies on tetanus of the last years are the main examples of this side of Abel's scientific activity. When a new field was started older interests were for the time cast aside, and all effort was devoted to mastering the new subject. It is, to say the least, extremely rare to find an investigator on his retirement at the age of seventy-five embark on the exploration of a field requiring totally different technique and methods from those with which he had been concerned for nearly half a century. Abel, however, did just this, and it is entirely in keeping with his fearless spirit, indomitable enthusiasm, youthful outlook and receptivity to new ideas, which all of his pupils recognized.

Several of Abel's publications appear under his name alone, but on most of them, especially in later years, we find the names of younger collaborators. Muirhead, Aldrich, Davis, Crawford and Taveau are the only collaborators up to the end of the epinephrine period; Ford, Rowntree, Barbour, Macht, Turner, Pincoffs, Rouiller, Kubota, Nagayama, Geiling, Bell and Wintersteiner were collaborators in the next period until retirement; the names of Evans, Hampil, Lee, Jonas, Firor and Chalian appear on one or another of his tetanus papers. The above list by no means includes all the many pupils or assistants who were in the laboratory at one time or another, many of whom never collaborated with Abel in his researches, but developed fields of interest of their own quite different from those of "the Professor." Important contributions on other topics appeared in an unbroken stream from his laboratory during the forty years of his directorship. Many of his pupils have occupied or now occupy important chairs of pharmacology, and even clinical medicine.

Abel found his greatest enjoyment in actively carrying on research with his own hands and became completely wrapped up in the major problem engaging his interest. In spite of his wide interests, he refused to allow himself to be side-tracked from his major objective of the time, which would appear to explain why he did not figure in the usual accompaniments of a successful scientist—medical societies, committees, boards, etc. In view of this rather limited activity outside of his laboratory and study, it is curious to see his intense interest in scientific journals and in the organization of national scientific societies. He served from 1896–1905 as associate editor of the *Journal of Experimental Medicine* when it was first established. He founded the *Journal of Biological Chemistry* with Herter in 1905 but withdrew from active editorship in 1909 to found *The Journal of Pharmacology and Experimental Therapeutics*, which he edited for twenty-three years. He issued the call and addressed the first gathering at which the American Society of Biological Chemists was founded, and formed the American Society for Pharmacology and Experimental Therapeutics. Here was something which he evidently regarded as of equal value to his research. Correspondence with many friends and acquaintances both in this country and in Europe was kept up during the whole long period of scientific activity. He knew how to take a true vacation, and dropped all work, and applied himself to this as intensely as he did to his scientific research.

The Universities of Michigan, Pittsburgh, Harvard, Yale, Lwow (Poland), Cambridge and Aberdeen conferred honorary degrees upon him. He was awarded the first Research Corporation prize, the Willard Gibbs, Conné and Kober medals and the medal of the Society of Apothecaries, London. A member of the National Academy of Sciences, honorary fellow or member of six American and fourteen foreign scientific societies, he received his last honor of Foreign Member of the Royal Society, London, the day of his death.

A mere description of the scientific work and numerous honors awarded him leaves one with a very incomplete and unsatisfactory picture of the man. The spirit of Abel was far greater than any of his scientific discoveries. He exercised a great influence on many pupils, assistants and others who came in contact with him during the course of his long scientific career; he served unconsciously as a very effective catalyst for the growth of scientific pharmacology in this country. He taught by example. None of the many who at one time or another were privileged to work in his laboratory failed to profit by his intense enthusiasm for his research, his youthful outlook on science, his tremendous optimism that the morrow would yield the coveted result, his fearlessness in engaging in difficult problems or in controversies, and above all the real simplicity of a very lovable man. None of these can forget the noon hour lunch table discussions where all the workers were thrown into intimate contact with "the Professor." Here the talk varied widely, sometimes largely shop talk, philosophical discussions, heated arguments, but above all a feeling of good fellowship with a certain amount of humor. The lift of the eyebrows and the merry twinkle in his eye as "the Professor" made some statement to provoke discussion will long be remembered by those who participated in these meetings. It was a special occasion when old pupils or assistants returned to lunch with the novitiates, to tell tales of the glories of the old days and anecdotes of the former conduct of their "Professor." Abel thoroughly enjoyed these luncheons and contributed largely to their success.

Despite his great absorption in his own major research problems of the moment, the door of his laboratory was always open to any worker in his laboratory or any former pupil or scientific friend. When one came for advice, one found a man who seemed to be working against time, but one who was quite willing to stop to give advice to a younger colleague.

His research activities seemed to fill and permeate his whole life—he regarded research as a sacred torch to be kept burning at all times. In one of his addresses he writes: "Greater even than the greatest discovery is it to keep open the way to future discovery. This can only be done when the investigator freely dares, moved by an inner propulsion, to attack problems not because they give promise of immediate value to the human race, but because they make an irresistible appeal by reason of an inner beauty . . . . In short, there should be in research work a cultural character, an artistic quality, elements that give to painting, music and poetry their high place in the life of man."

A truly great international figure has passed. His many pupils, friends and acquaintances can not help but feel the loss, but can be reconciled to it by his long life of accomplishments and by the fact that he "died in harness" as he had wished.

The words of Socrates, which he once used to describe his old teacher Carl Ludwig, might well be said of John J. Abel. "A man whose desires are drawn towards knowledge in every form and who is therefore absorbed in the pleasures of the soul—one who is harmoniously constituted, who is not covetous or mean, or a boaster or a coward and can never therefore be unjust or hard in his dealings—he has no secret corner of meanness and is a searcher after and lover of the truth in all things."

E. K. Marshall, Jr.

THE WILLARD GIBBS LECTURE

Your speaker to-day is one who is primarily a worker in the field of experimental medicine; a chemist, if at all, only in so far as an imperfect mastery of your science became necessary for the solution of physiological and pharmacological problems that could not be undertaken or even formulated if their chemical aspects were to be ignored. Under the circumstances I can but feel a sense of deep unworthiness in venturing to address an audience in which are gathered so many distinguished representatives of your noble science. I am highly appreciative of the signal honor conferred upon me by the board of award of the Chicago Section of the American Chemical Society in the bestowal of the Willard Gibbs Medal and I beg the members of the board to believe that I am duly grateful to them.

There exists in our day an essential unity of outlook and interest among the majority of professional chemists, biologists and medical men in respect to the physical and chemical aspects of life. This unity of interest and unanimity of opinion in respect to the applicability of the laws of physics and chemistry to the elucidation of vital processes have their origin far in the past and date from a time long before chemistry had attained to its present dignity as an independent science. It is not my purpose to attempt to record even briefly the history of chemistry or that of medicine, subjects that have been so well treated by many learned men of both professions, but I would ask your forbearance toward an imperfect sketch of the points of contact between your professional ancestors and mine. I leave out of consideration here any reference to such contacts in the ancient or later alchemical periods, or to Arabian science in Western Europe, further than to remark that alchemy, which at its best combined far-reaching metaphysical speculations with a crude experimental chemistry, had, as one of its several aims, not alone the transmutation of the baser metals into gold, thus abolishing that "great disease, poverty," but also the cure of fleshly ills and the gift of "perfect health and length of days." Certainly the search throughout the long alchemical period for the "elixir of immortal health" supports the claim for this medical aspect of alchemy, even without proofs on the literary side.

To-day the modern alchemists among you have, in a most remarkable manner and in a most concrete way, actually realized the age-long dream of your predecessors. Paneth well describes the chemical and the medical actions of the modern philosophers' stone, radium, in the following words:

> Thus we see that in a certain sense radium possesses the first and principal property ascribed to the philosophers' stone: it has the power of transmuting elements, although not of producing gold. And, oddly enough, even in respect to the second property which was ascribed to the philosophers' stone radium seems to have gotten something from its fabulous predecessor: it is a very valuable aid in the treatment of some

severe diseases, although not a perfect remedy for every illness. So that to a certain degree the radium rays really produce the two very different effects of the philosophers' stone, transmutation and healing.

Even in the later alchemical period the older professions of medicine and pharmacy furnished opportunity, though often grudgingly given, for the development of chemistry, and they may justly be said to have been the parents of modern chemistry. Throughout this period, as in ancient times, there existed a large number of industries, such as the metallurgical industries, enamel- and glass-making, painting, brewing and wine-making, to give only a few examples. But the practice of these ancient arts could not lead to the development of a chemical science as long as the true character of organic prinicples of the elements and their compounds remained unknown. Especially close was the connection between chemistry and medicine in the days of the iatro- or physician-chemists of the sixteenth and seventeenth centuries. In the first half of the sixteenth century, at a time when the crude chemistry of that day was still entangled with the traditions and even downright impostures of alchemy, there appeared an extraordinary man, Paracelsus (1493–1541), "the very incarnation of the spirit of revolt" (Osler), who must indeed be regarded as one of our ancestors in both branches of learning. Remember that in his day medicine was already a long established and powerful profession and this bold innovator in chemical physiology, pathology and pharmacology, who heaped scorn on Galen and Avicenna, very naturally aroused the hostility of many of the leading physicians of the day.

He is the most notable figure among the earlier physician-chemists and, in the words of the historian E. v.

Meyer, to him belongs incontestably the credit of fusing chemistry and medicine in the first half of the sixteenth century, of forcing both into new paths and of freeing chemistry from the shackles of alchemy. Up to our time some of his writings have appeared in no less than five hundred editions. He declared it the true purpose of chemistry not to be the making of gold but the preparation of medicines. "Alchemy is neither to make gold nor silver: its use is to make the supreme sciences and to direct them against disease." This too narrow a conception of the science was soon to be broadened out by the iatro-chemists of the century and more, following his death. He stated clearly that the processes of the animal organism are chemical in their nature and that health is a function of, or dependent on, the composition of the juices and tissues of the body. Even in regard to the rôle played by air in respiration, I find Robert Boyle, more than a century later, citing him in support of his own views, as saying "that as the stomach concicts meat and makes part of it useful to the body, rejecting the other part, so the lungs consume part of the air and proscribe the rest." Medical historians have occupied themselves more with an analysis of the medical achievements of Paracelsus, the Luther of medicine (Osler), than with his chemical discoveries, and his significance is perhaps greater for medicine than for chemistry. He is certainly of great importance in the history of chemical therapeutics and pharmacology. Sudhoff says of his work in these fields:

> He was the first one to show how to separate the active principles from drugs and to use them in tinctures and extracts. He made important discoveries in chemistry: zinc, the various compounds of mercury, calomel, flowers of sulphur, among others, and he was a strong advocate of the use of preparations

of iron and antimony. In practical pharmacy he has perhaps had a greater reputation for the introduction of the tincture of opium—"labdanum" or laudunum—with which he effected marvelous cures and the use of which he had probably learned in the East (Osler).

Naturally, I can not pause to give more than a very incomplete sketch of this innovator whom I here mention because of his constant insistence that physiological processes are chemical processes and that drugs by their chemical properties can bring about favorable chemical alterations in a diseased body.

It has for many years been a custom with me to characterize the vital importance of certain defensive substances and the products of internal secretion present in our bodies in the aphorism: We are walking drug stores. On reading Sudhoff's reprint of the *Labyrinthus medicorum errantium* of Paracelsus (1538), a little meaty classic, I was delighted to find that four centuries ago our author had already elaborated this idea at length:

> Also in human beings there is a natural apothecary in which are found all things as in the world (the macrocosm), good and bad, simples and composites, however they be named. . . . As the outer world, the macrocosm, contains visible pharmacies and visible physicians, so in the microcosm, that is to say, in the human being, there is present an invisible pharmacy and an invisible physician who produces, prescribes, dispenses and administers suitable remedies as occasion demands. . . . Let it be known to all men then that had not God created and placed in the bodies of men natural remedies and a natural physician, then, notwithstanding all the efforts of our physicians, not a single creature of earth would remain alive.

In this quite modern fashion Paracelsus here restates the enduring canon of Hippocrates—*Νούσων φύσιες* *ιατροί*—better known to us in its later paraphrased form as *vis medicatrix naturae.* Professor Max Neuburger, the learned professor of the history of medicine at Vienna, truly remarks that the problem of nature's healing power is probably the most weighty of all that have engaged the thoughts of physicians during thousands of years.

The iatro-chemists were the physiological chemists of their day. They possessed the highest scientific and humanistic culture of the time and historians of science agree in the opinion that chemistry profited greatly by passing into their hands. The study of their attempts to elucidate the phenomena of life and the chemical alterations associated with disease is a fascinating one. The physiologists, biochemists and pharmacologists of our day have at their disposal a great wealth of discovery in the more exact sciences—the gifts of the intervening centuries, but even with all this assistance modern iatro-chemists still find themselves trying, though confidently, to hack their way out of the jungle so boldly and gaily entered by their predecessors nearly three hundred years ago.

Naturally you will call to mind also, as every one must, the iatro-physicists of this period. It is thought that the physiological speculations of Decartes, as given in his treatise *De Homine*, had a great influence in turning men's minds to the possibility of basing physiology on physics and chemistry, but there is little doubt that the teachings and discoveries of Harvey had the greater influence on the development of iatro-physics. This phase of science, however, lies outside our present discussion.

I must content myself with recalling to your minds a few of the names of the iatro-chemists of this period—van Helmont and Sylvius, of whom Michael Foster, with a first-hand knowledge of their writings and those of their pupils, has given such an enlightened and sympathetic account in his Lane Lectures on the History of Physiology in the sixteenth and seventeenth centuries—Tachenius, Willis, Mayow, Lémery, Hooke, Peyer, Brunner and many others of this time might be named who forwarded both chemistry and medicine in the latter half of the sixteenth and well on into the seventeenth century.

One great man, Robert Boyle, nobleman of wealth, stands out prominently in the seventeenth century, one whose name is revered alike by physicists, chemists and medical investigators. This greatest chemist of his day, though not trained as a medical man, nevertheless, says one of his biographers, Thomas Birch, "went very accurately through all the parts of Physic." A study of his writings furnishes ample evidence of his acquaintance with medicine and the medical theories of his day. This great man even occupied himself with therapeutical questions and published "collections of choice and safe remedies, for the most part simple and easily prepared, very useful in families and fitted for the service of country people." The historian Neuburger in his scholarly treatise entitled "Die Lehre von der Heilkraft der Natur im Wandel der Zeiten" devotes several pages to Boyle's views in regard to the respective rôles of "nature" and the physician "in restoring the distempered body to its pristine state of health," and states the Boyle's opinions on these matters were not without value and precipitated many controversies.

Boyle was particularly fascinated by that fundamental problem—the nature of the respiratory process—of which he says that "it is a subject of that difficulty to be explained and yet of that importance to human life that I shall not regret the trouble my experiments have caused me if they are found in any degree serviceable to the purpose for which they were designed." I have been surprised to learn, in my study of his writings, with what ardor Boyle pursued the effects of diminished air pressure, with his improved Guericke "pneumatic machine," on a great variety of animals. Bees, flies, butterflies, caterpillars, humming birds, sparrows, larks, mice, fishes, eels, unborn puppies, all served as objects of experiment in the study of this great problem which was to occupy the attention of his successors down to our day. At a time when, in spite of some earlier approaches to the truth, it was still generally held that the sole purpose of breathing is to cool the blood, Boyle's experiments forced upon him the conviction expressed as follows:

> Without denying that the inspired and expired air may be sometimes very useful by condensing and cooling the blood that passes through the lungs, I hold that the depuration of the blood in that passage is not only one of the ordinary, but one of the principal uses of that passage. But I am also apt to suspect that the air doth something else in respiration which hath not yet been sufficiently explained.

The last sentence suggests that Boyle may have had a prevision, years before the discovery of oxygen, that this "something else" is the respiration of the tissues themselves.

After citing the opinion of Paracelsus, an opinion not based on experimental evidence, that "the lungs consume part of the inspired air and proscribe the rest," Boyle, on the basis of his own extensive experimentation, makes the prophetic statement:

> It seems we may suppose that there is in the air a little vital quintessence, if I may so call it, which serves to the refreshment and restauration of our vital spirits, for which use

> the grosser and incomparably greater part of the air being unserviceable, it need not seem strange that an animal stands in need of almost incessantly drawing in air.

The "vital quintessence" is of course the later oxygen of Priestley and Scheele.

All of his experiments lead him to support

> the theory of Moebius that the genuine use of respiration is the ventilation not of heart but of the blood in its passage through the lungs, in which it is disburthened of those excrementitious steams proceeding for the most part from the superfluous serosities of the blood.

Truly, Boyle could touch no subject without leaving his mark on it. For example, the respiration of fishes, "being animals without lungs," also excited his curiosity, and he "thinks it not altogether absurd to say that their gills seem somewhat analogous (as to their use) to lungs." His experiments with the air pump had taught him that there is "wont to lurk in water many little parcels of interspersed air, whereof it is not impossible that fishes may make some use, either by separating it when they strain the water through their gills or in some other way."

Time and occasion permit only of a glance at a few other great peaks in the panorama that we are so hastily surveying. Let us pass at once from Boyle to Lavoisier, another immortal, one of the greatest among the founders of your science, often, indeed, called the creator of modern chemistry, but also one who made fundamental contributions to the theory of respiration. He mistakenly held that combustion occurs only in the lungs. Thus, in the well-known joint memoir with the great mathematician Laplace, published in 1780, the conclusion is arrived at:

> Respiration is therefore a combustion, slow, it is true, but otherwise perfectly similar to the combustion of charcoal. It

takes place in the interior of the lungs without giving rise to sensible light because the matter of the fire (the caloric), as soon as it is set free, is forthwith absorbed by the humidity of these organs. The heat developed by this combustion is communicated to the blood which is traversing the lungs, and from the lungs is distributed over the whole animal system (Foster's translation).

In spite, however, that the place where oxidation occurs is erroneously inferred to be in the lungs only rather than in the hidden recesses of the tissues of the body, Lavoisier is nevertheless the first chemist or physiologist to *prove* that the respiration exchange is the result of a combustion and he was the first to make quantitative measurements during respiration of the intake of the "respirable part" of the atmosphere (the oxygen of Scheele and Priestley, later so named by Lavoisier) and of the output of the "aeriform calcic acid" (the carbon dioxide or "fixed air" of Black). It is quite comprehensible that the creator of gravimetric analysis and the discoverer of the principle of the conservation of mass in chemical operations should have made this quantitative experiment in animal physiology.

There is not time while we are on this subject of the respiration to consider the work of Joseph Black, Robert Hooke, Richard Lower and others of that time. The physician-chemist John Mayow (1643–79), however, can not be passed over without a word. Like Boyle, Mayow saw clearly that respiration is supported, not by the air as a whole "but by the more active and subtle part of it, the spiritus nitroaerius," or spiritus igneo-aerius, and both by experiment and inference demonstrated the analogy between respiration and combustion. The increase of weight attending the calcination of metals he declares also as due to the absorption of the *spiritus* of the air. Mayow had sound notions in regard to the relation between increased muscular work and increased respiration a full century before Lavoisier and Priestley, but unfortunately, in conformity with the conceptions of his day, he states that his nitro-aerial spirit is separated from the blood in the ventricles of the brain and passes thence in supposed nerve tubules to the muscles where it combines with "sulphur," in consequence of which union muscular contraction results. The numerous and ingenious experiments of Priestley (1733-1804), one of the last phlogistonists, on respiration, combustion and calcination, as detailed in his memorable treatise, Experiments and Observations on Different Kinds of Air (1774), must also be passed over.

The latter part of the eighteenth and the first quarter of the nineteenth century were days of great discoveries of inestimable value to mankind, discoveries that inevitably and finally raised chemistry to the status of a great science, a younger sister of physics, though in our day the exponents of the older science claim, and perhaps justly so, that your science will ultimately be incorporated into the body of physics and that instead of atomists you will become mathematical energeticists, dealing primarily with protons and electrons and quanta and other concepts of their wizardry.

In this period the elemental or composite character of the various "airs" that had so puzzled the men of Boyle's day was determined: oxygen, nitrogen, hydrogen, methane and the composition of water and carbon dioxide came to be known. The chemical balance came into daily use. New elements were discovered and new inorganic compounds were made. An increasingly large number of organic compounds were being isolated as definite chemical individuals from a variety of vegetable products and from the tissues and secretions of man and animals. In this period falls the discovery and isolation of some very important products of animal metabolism, as urea (1773), uric acid (1776), allantoin (1800), cystine (1810), creatine (1835), glycerin (1779) and other chemical principles, the study of which was later to exert so great an influence on the development of synthetic chemistry. In this period also the pharmacist-chemists isolated quinine from the cinchona bark, that beneficient febrifuge (*Pulvis febrifugus orbis americani*) introduced into Europe from the new world nearly two centuries before; that gift of the gods, the juice of the poppy, was made to yield in crystalline form its analgesic morphine and its less valuable sister alkaloid, codeine (1803–1823). The seeds of the strychnos tree yielded up strychnine and brucine; hellebore and cevadilla seeds, veratrine; ipecacuanha, emetine; tobacco, nicotine; and a little later atropine was isolated from belladonna. A discovery of fundamental importance for chemistry, medicine and the arts was Faraday's isolation of benzene from gas distillation residues in 1825. Of his "bicarburet of hydrogen," by which name the new hydrocarbon was designated, Thorpe wrote a quarter of a century ago: "the work which has accumulated around this single substance during the 75 years which have elapsed since it has been known constitutes one of the most astonishing records of intellectual and industrial activity of which history has any record."

But the first quarter of the nineteenth century was a period when men did not solely occupy themselves with the isolation of elements and new compounds but, as in the quarter of a century that we have just lived through, minds of a high order also developed theoretical principles. Years after the enunciation of Boyle's Law and more or less coincidently with a host of brilliant discoveries of the very greatest importance in various departments of physics and in applied mathematics appeared Avogadro's law and Dalton's atomic theory. I need only recall to your minds the names of some of the greatest investigators in physics of this era, as Faraday, Ampère, "the Napoleon of electricity," Oersted, Ohm, and preceding them, Galvani, Volta and Coulomb among those who laid the the foundations of electro-dynamics; of Thomas Young and Fresnel in the field of light, of the great Carnot and of Fourier in that of heat, of Gay-Lussac and Mariotte in the field of the gases, of Dulong and Petit, who showed that the atoms of elementary substances have the same capacity for heat. The important discovery that substances having an identical elementary composition may yet differ in their chemical or physical properties or both—isomerism—also falls in this period (1823), as also the enunciation by Grotthus (1818) of the fundamental principle underlying all chemical processes involving the absorption of light energy. It was indeed a remarkable period. Science and biology have been benefited as much or even more in consequence of these discoveries in physics of a century ago than by the chemical additions to knowledge that were coincidently made.

The formulation of such general statements capable of mathematical treatment is, as is known to all, the highest aim and the ultimate goal of the scientist's activity. We, who deal with the chemical and physical complexities of living organs, where the variables are not only very numerous but also where the dependence of each of these variables on one or more of the others must be taken into account, can but regard the full attainment of these highest aims of biological science as something that lies in the far future.

It should be stated, however, that many special topics in the broad field of plant and animal physiology are already capable of quantitative treatment, and many of my colleagues will feel that I have understated our case. They will call to mind investigations in relation to vision, muscle contraction, the nature of the nerve impulses, the neutrality mechanism, respiration and metabolism, to cite only a few examples from physiology to which quantitative methods are applicable.

But let us trace still further the relationship between chemistry, biology and medicine. The character of this relationship is now becoming reversed. No longer are the devotees of the biological and medical sciences foster-parents or foster-brothers of yours as in the old days. It is we who are now dependent in a large part of our work upon you and your fellow scientists, the physicists.

The reason for this change in relationship is not far to seek. It lies primarily, as stated above, in the fact that your science has devised many more diverse and accurate methods of measurement.

THE ERA OF CHEMICAL SYNTHESIS

Hardly had the first quarter of the nineteenth century passed when new developments arose, more especially in organic chemistry, that were destined to push forward in an immeasurable degree not only man's ultimate conquest of nature but also his ability to control or conquer disease and to prolong life, and hence, in so far as this may contribute to that end, to increase the sum of human happiness. The new era that has brought priceless gifts to mankind is the era of chemical synthesis. In conformity with my plan of emphasizing the importance, for the biological sciences and medicine, of only the great achievements of your science in this era I must limit myself to a brief survey of the discoveries of a few among the great men whose work happens to be of especial significance for these sciences. Let me begin with Liebig, whose contacts with medicine came through pharmacy, to which profession he was apprenticed in his early youth, and who later continued his chemical studies in Paris under Gay-Lussac. In his "Organic Chemistry in its Applications to Physiology and Pathology," published in 1842, after stating that "the great physicians who lived toward the end of the seventeenth century were the founders of chemistry and the only philosophers acquainted with it," and deploring the fact that "modern chemistry, with all its discoveries, has performed but slender services for physiology and pathology," he gives expression to his belief that:

> The most beautiful and elevated problem for the human intellect, the discovery of the laws of vitality, can not be resolved—nay, can not even be imagined—without an accurate knowledge of chemical forces: of those forces which do not act at sensible distances; which are manifested in the same way as those ultimate causes by which the vital phenomena are determined; and which are invariably found active whenever dissimilar substances come into contact. . . . Before the time of Lavoisier, Scheele and Priestley, chemistry was not more closely related to physics than she is now to physiology. At the present day chemistry is so fused, as it were, into physics that it would be a difficult matter to draw the line between them distinctly. The connection between chemistry and physiology is the same and in another century it will be found impossible to separate them.

After this confession of faith by Liebig we can no longer wonder that this great chemist concerned himself during many fruitful years with numberless questions touching the life processes of plants and animals; that he should be remembered to-day not only for his lasting achievements in organic and in technical chemistry, for having been a great teacher and inspirer of young men, but also as the founder of agricultural chemistry and as the most notable among the modern founders of biological chemistry since Lavoisier.

You all know that to the beautiful friendship which existed between him and the great Wöhler, unique in the history of science, and to the fortunate collaboration of the two in research, we owe some of the very greatest contributions ever made to the subjects now under consideration. Friedrich Wöhler studied medicine as a young man, taking his degree in medicine and surgery at Heidelberg in September, 1823. While a lad at the Gymnasium, he devoted himself passionately to chemical experimentation and to the collection and study of mineralogical specimens, much to the neglect of his school subjects. At Heidelberg he came into close contact with the chemist Gmelin and with the physiologist Tiedemann. Before he had taken his degree the young Wöhler had already completed four investigations dealing with selenium and with cyanic compounds and the year (1824) after his graduation he published a very comprehensive study, originally intended for a graduation thesis, of the excretion in the urine of a very large number of substances, including iodine, carbonates of the alkalis, saltpeter and other inorganic compounds; numerous organic acids, including benzoic acid, substances that color the urine, as indigo and rhubarb, and such as impart an odor to this secretion, as turpentine and asparagus. This paper, entitled "Experiments on the Passage of Substances into the Urine," was awarded a prize by the medical faculty of Heidelberg, and can be read to-day with profit by physiologists and pharmacologists, not along for its numerous references to the work of other investigators of that time but also because of the wonderful acumen with which the youthful investigator discusses the true function of the kidneys and the cause of the acidity of the urine. He sums up his deductions in regard to the kidney in the following passage:

> The kidneys are organs that serve the purpose of secreting a fluid composed in part of such substances as pass in the unassimilated condition into the blood, being incapable of serving as replacers of bodily constituents, and in part of such as are produced during digestion and during the interchanges of material in the animal body (intermediary metabolism) or which are finally thrown off as being of no further use in these interchanges; they are therefore organs that serve to maintain the composition of the blood in a state necessary to the maintenance of life, without themselves producing any new substance whatever.

Pharmacologists of a later day were to show that Wöhler's "physiological deductions" needed to be modified in that the kidneys have a definite though limited power of synthesis and physiologists have also proved that, in addition to the kidneys, the respiratory function serves also to maintain the composition of the blood and tissue juices in the chemical equilibrium necessary to life.

Fortunately for both medicine and chemistry, Liebig and Wöhler in 1837 elected to attack the problem of the chemical constitution of that important metabolite, uric acid, first isolated by Scheele from urinary calculi in 1776, which up to that time had been of interest only to medical investigators and practitioners because of its connection with gout. As A. W. Hofmann so aptly remarks, this constituent of our body has shown itself to be a very protean among organic chemicals.

The results obtained by these two great investigators in their study of this acid and of the more than sixteen entirely new derivatives which they were able to prepare from it, as also of the many previously known substances that were encountered by them during their oxidation and reduction experiments in this field, have been described in detail by A. W. Hofmann and have also been outlined in a fascinating manner by T. E. Thorpe in his essays on historical chemistry. These memorable researches on uric acid excited the highest admiration of the chemists of the day, and I need not comment further on their significance for organic chemistry or for the future development of chemical physiology.

I need not comment either at length on the great consequences for organic chemistry and for medicine that flowed from the collaboration of the pair in their researches on the nature of the oil of bitter almonds, suggested by Wöhler to Liebig in 1832 as a suitable subject for a joint research. It must have been something akin to inspiration, says Thorpe, that led Wöhler to suggest the subject. I have often wondered whether the germ of the idea could not be traced to Wöhler's wide knowledge of medicinal substances. Oil of bitter almonds had long been in use as a domestic remedy, and Wöhler, who, in addition to his professorship at Göttingen, also held the post of supervisor of pharmacies for the Kingdom of Hanover, must have been well aware of the medicinal properties of the oil. The discovery of the compound radical benzoyl (and of benzoyl chloride) and the proof that numerous correlated bodies could be grouped around it were epoch-making in their far-reaching consequences and will always remain, historians of science are agreed, one of the greatest achievements in organic chemistry.

Wöhler's discovery in 1842, made also independently by Alexander Ure, of Edinburgh, in 1841, that ingested benzoic acid is paired in the animal organism with glycocoll, or amino-acetic acid, and excreted as hippuric acid, which, from a purely chemical point of view, may be regarded as only a minor addition to the "benzoyl" edifice, became nevertheless of the greatest significance for physiology and pharmacology as constituting the first demonstration that the animal organism has the power to combine chemically dissimilar substances into new compounds—a capacity for synthesis that up to that time was believed to be confined solely to plants. This discovery naturally led to wider views in respect to the nature of intermediary metabolic processes. Since 1842, biochemists and pharmacologist have discovered innumerable other instances of the very considerable synthetic power with which animal tissues are endowed and the conception no longer excites wonder by its novelty.

The significance of Wöhler's discovery in 1828 of the artificial formation of urea by the molecular transformation of ammonium cyanate is known to every freshman in our schools of chemistry and medicine. This discovery, the significance of which was fully realized by the young Wöhler, effected a revolution in the ideas of men and showed that the organic substances present in bodies of animals and in plants can not be the products of a mysterious vital force but are subject to the same laws as those that are encountered by the chemist in his laboratory while occupying himself with bodies of purely inorganic origin.

Of the chemists of the period under consideration who were intensely interested in the chemical phenomena presented by plants and animals, I can refer here only to the great French chemist, Jean Baptiste André Dumas, the contemporary of Wöhler and Liebig. At the age of eighteen, the young pharmacist detected the presence of the newly discovered iodine in burnt sponge, a substance employed at the time in treatment of goiter, and later worked in conjunction with the physiologist Prévost on the active principle of digitalis, on the transfusion of blood, on the seat of formation of urea in the body, on the chemical changes accompanying the embryonic development of the chick and many other chemico-physiological question. Naturally, even a man of Dumas' ability could not, at that time, make any significant advance in our knowledge in his too brief excursions into these difficult fields. He and Prévost were, however, the first, as far as I am aware, to demonstrate by experiments on nephrectomized animals that urea is not formed in our kidneys but elsewhere in the system, as they were still able to detect it in the blood of the nephrectomized animals.

In his later years, long after he had won his place among the great chemists of his time, Dumas returned, says Thorpe, to many of the chemico-physiological problems with which, in association with Prévost, he had begun his career as an investigator. In this later period fall his studies with Boussaingault on the formation of fat in the animal body and his own studies on the origin of bees'-wax. He proved that bees, even when fed exclusively on sugar, still retained the power of producing wax, contrary to the opinion of his predecessors, who believed that the bee secretes the wax while extracting the honey from flowers and that there is no necessary connection between the two processes. The nature of alcoholic fermentation, the treatment of the silkworm disease and methods for destroying the vine pest, *Phylloxera vastatrix*, were among the later interests in the life of this gifted and many-sided man.

Having unfortunately little leisure for the pursuit of historical studies, I am fully conscious of the fact that I have given you a very sketchy outline of the points of contact between chemistry and medicine from Paracelsus to the time of Liebig, Wöhler and Dumas. The problem of animal respiration we had left in the state in which it was formulated by Lavoisier. But even after Spallanzani and others had shown early in the nineteenth century that consumption of oxygen takes place in the tissues themselves rather than in the blood, a true solution of the problem could not be arrived at without quantitative data in regard to the oxygen and carbon dioxide content of both arterial and venous blood. Here a non-medical investigator, Gustav Magnus, professor of physics at Berlin, came to the rescue of physiology in 1837. By means of the mercurial air pump he was able to liberate oxygen and carbon dioxide from both arterial and venous bloods and to show that the percentage of oxygen is greater in arterial blood while that of carbon dioxide is greater in venous blood. These determinations by Magnus were afterwards amplified by the physiologists Pflueger, Ludwig and his pupils, among whom was Lothar Meyer, later to win distinction in pure chemistry, and their work, in conjunction with the labors of many other investigators during the last half of the nineteenth century, has given us a theory of both internal and external respiration which the science of our day has only been able to expand but not to overthrow. The historian Fielding H. Garrison pithily remarks that "the development of the physiology of respiration from Borelli to Magnus was almost exclusively the work of three mathematicians, two physicists and five chemists."

In respect to the further developments in regard to both internal and external respiration, I cannot do better than transcribe the concise description by one of your own guild, Professor W. Mansfield Clark:

> In all science there are few developments as beautiful as those which have given us the precise knowledge of the blood equilibria. There have been found: a quantitative relation

between the iron of the blood pigment and the oxygen combining capacity; quantitative data for the equilibria between partial oxygen tensions and degree of oxygen saturation of haemoglobin; preliminary data on the Donnan equilibrium between the oxygen-carrying blood pigment, trapped within the semipermeable menbrane of the red cell, and the plasma; exact relations for the bicarbonate equilibria of the plasma and the acid-base properties of the oxygen carrier; the mechanisms for the maintenance of constant hydrogen-ion concentration of the blood and the control of lung ventilation by the activation of a nerve center called the respiratory center. Of this Haldane says, "A rise of 0.2 per cent. or 1.5 mm. in the CO_2 pressure of the alveolar air and arterial blood causes an increase of about 100 per cent. in the resting alveolar ventilation. The outstanding delicacy of the regulation of blood reaction is thus evident. No existing physical or chemical method of discriminating differences in reaction approaches in delicacy the physiological reaction."

The prophecy of Liebig has been realized. In our day chemistry and physics have become fused into the very structure of the biological and medical sciences. But despite the fact that chemists during the past seventy-five years have necessarily concerned themselves largely with the development of their science along inorganic, synthetic, physicochemical, quantitative and, in recent years, more and more along electrochemical and mathematical lines, they have nevertheless, as in earlier times, continued to take an interest in the chemistry of life processes. It would too greatly lengthen this paper to give even a brief outline of the hundreds of contributions that have been made by the chemists of all countries to biochemistry since 1860 or thereabouts.

An outstanding example or two of the contributions of chemists to biology from that time to ours must suffice. And here there naturally comes first to mind the name of

Emil Fischer, one of the very greatest experimental chemists of all time, whose outstanding contributions toward the solution of the most difficult and important among biochemical problems will always be looked upon with veneration by those who are conversant with the researches of this genius and have a first-hand knowledge of these difficult subjects. His researches on the chemical structure of the sugars and on the very specific ferments that liberate them from their derivatives, and on the structure of the proteins and their primary and secondary cleavage products effected a revolution in our chemical concepts of these products of vital activity and opened up new vistas in regard to biochemical processes. An analysis and a description of his monumental discoveries in regard to the members of the purine group and their allies, as found in both animals and plants, would carry us back to Scheele's discovery of uric acid in urinary calculi and then to a consideration of the earlier discoveries of his predecessors, Wöhler, Liebig, Fourcroy, Grimaux, Baeyer, Strecker, Horbaczewski and others. Suffice it to say that the contributions of Emil Fischer surpass in their ultimate significance for chemical physiology those ever made by any other man in the entire history of biological and medical science.

By substitution, degradation and especially synthesis, he established the genetic relationship between uric acid, xanthine and hypoxanthine and their multitudinous substitution products and showed that they may all be considered as derived from a substance $C_5H_4N_4$, for which he proposed the name purine (from *purum* and *uricum*) and which, although purely hypothetical at the time, he afterwards succeeded in synthesizing. In the field of the sugars, with the aid of phenylhydrazine, which he himself had discovered at the very outset of his scientific career (1875), he was able to isolate in pure form not only the few then known natural monosaccharides, but a host of others whose existence he predicted from the van't Hoff theory of the asymmetric carbon atom and which he synthesized by the skillful application of a number of general reactions, and in this way established the structures and configurational relationships of this very important group of substances. He also studied in detail and clarified the chemistry of the glucosides, their structure and spatial configurations and especially their behavior towards enzymes. The ester method for the separation of amino acids which he devised enabled him to determine much more nearly quantitatively than had hitherto been possible the composition of the complex mixtures resulting from the hydrolysis of proteins, and led to the discovery of several still unknown amino acids in such mixtures. These amino acids he succeeded in condensing with each other in amide-like union to substances which he called peptides and which, as the number of the amino acid residues of which they were made up was increased, approached closer and closer in physical and chemical properties to the peptones obtained from natural products. Similarly, he found that the tannins are glucosides of ester-like combinations, "depsides" (from δέψειν, to tan), of phenolcarboxylic acids joined to each other through a phenolic hydroxyl group of the one and the carboxyl group of the other. As in his protein work, after having determined the nature of the component units of the natural products, he synthesized numerous compounds more and more resembling these natural products in their chemical reactions and physical properties.

As I am noting here more particularly the contributions of certain leaders among so-called pure chemists toward the elucidation of the chemical occurrences in living things, rather than those of biochemists, pharmacologists and physiologists, significant as these have been, it is only natural that I should refer to the work of Willstätter, A. v. Baeyer's distinguished pupil. You are all familiar with the brilliant researches of this master and his pupils on the chemical nature of chlorophyll and with their success in isolating in crystalline form the chemical individuals that had originally been grouped under that comprehensive term. It has been said that if it can be assumed that one thing is of more importance than others, then chlorophyll is undoubtedly the most important and the most indispensable of all things. The epigram contains a larger element of truth than is usually inherent in pithy sayings. Certainly the chloroplast of the green leaf, one of the most wonderful of all chemical laboratories, activated, as it is, by the sun's radiations, taken into consideration with the chemical activities of the other plant physiological mechanisms, offers problems that challenge the highest skill of the chemist and biophysicist. And here in this broad field of photo-synthesis we find pure chemists, plant chemists and physicists combining forces in our day as never before, in the solution of problems of the greatest significance, both from a theoretical and a practical point of view.

Willstätter and his collaborators, building on the foundations laid by earler workers, Hoppe-Seyler, Gautier and others in the eighties of the last century, v. Nencki, Küster and others since 1900, have finally given us a clear picture of the chemical relationship that exists between haemoglobin, the respiratory pigment of our blood, and the assimilatory chlorophyll pigments of plants. The relationship is of philosophical as well as bio-chemical interest. Both classes of pigments have essentially a comparable structure, the basic complex of each, called aetioporphyrin by Willstätter, being a compound consisting of four substituted pyrrole nuclei united through two carbon atoms. The blood pigment contains iron and the plant pigment magnesium in combination, partly through normal and partly through secondary valences, with the nitrogen atoms of the pyrrole groups in the aetioporphyrin complex. On degradation the two pigments yield the red, metal-free porphyrins which are broken down by oxidation into the anhydride or imide of haematinic acid and this by loss of carbon dioxide gives methylethylmaleinimide.

A second field of the greatest importance to both plant and animal physiology which has been greatly advanced by Willstätter's researches is that of the enzymes or organic catalysts. These products of vital activity, in contrast to inorganic catalysts, such as the acids, the various elements and metallic compounds employed in our laboratories, are so highly specific in their action that a particular biose, as milk sugar, cane or malt sugar, for example, is only capable of being hydrolyzed by a special enzyme, the only one, indeed, that is capable of effecting its hydrolysis. By devising new and ingenious modifications of a method long employed in biochemistry, that of adsorption, this investigator has been able to separate from their mixtures, and to obtain in a high degree of purity, many enzymes and to differentiate, for example, two such closely related sugar-splitting enzymes as saccharase and maltase. The skilful employment of metal hydroxide gels, more particularly the various structural modifications of aluminium hydroxide, enabled Willstätter to effect these extraordinary separations of enzymes from the various impurities and the activating and inhibiting substances that are always associated with them in the naturally occurring mixtures with which we have to deal. The new methods may truly be described as selective adsorption methods, as has been done by Willstätter, and it is interesting to note, as he points out, that this selective adsorption is determined, not by the degree of dispersion or other physical state of the adsorbing compound, but rather by its chemical structure.

The researches of a century have shown how very numerous are the specifically acting enzymes that are present wherever protoplasm functions, whether in single-celled organisms or in the more intricate structures of the higher plants and animals, and how varied are the chemical operations whose entire course, that is to say, whose rates of reaction and elaborated products are determined by these unique agents of vital activity. Admittedly, the further elucidation by chemists of both the dynamics and the organic structural problems here presented will yield results of the greatest significance both for chemical science and for the better comprehension of the life processes. For your consideration as chemists, I shall conclude my remarks on this aspect of biochemistry by the citation of a passage from a recent address of Willstätter on "New Methods in Enzyme Research":

> Of the characteristics of enzymes only the structural chemical and probably also the stereochemical specificity are uninfluenced and constant. The sugar-splitting enzymes show the strictest specificity in both respects, as regards chemical constitution and arrangement in space; the fat-splitting enzymes have a wider range of activity on structurally different, ester-like substrates. But they appear to be more finely differentiated in their stereochemical specificity. If we compare the lipases of the pancreas, the liver, the stomach and the

fungi as to their action on the same racemic substrate we find all different from each other in their selective action, preferring, now in the one case, now in the other, the d- or the l-form, if only we use a sufficiently large number of the racemic esters, as, for example, of the mandelic acid group. This configurational specificity can so far be considered as an enzyme constant. Thus far there has been observed no case where the influence of foreign bodies determines the direction of the rotation of the preferred compound.

Even a rapid survey of the many other fields in which chemists and biologists have an interest in common would transcend the limits that time and a consideration for your patience impose upon me. There remain, for example, the problems that arise in connection with the nature of the oxidation and reduction processes that are carried out in our bodies at 37.5° C.—problems that stand in the closest theoretical relationship to the efforts of your fellow chemists to explain the true mechanism of even the simplest of oxidations, as that of carbon monoxide to carbon dioxide. My personal interest in this most fundamental of biochemical problems stands in inverse ratio to my ability to treat it adequately, even were there time at my disposal to do so. Fortunately I can refer you to the concise and elegant outline of this subject that was given us in Baltimore two years ago by Professor Stieglitz in his Dohme lectures. This field, I am happy to say, is being tilled intensively by the younger physical, organic and biochemists of the day. The earlier experimental observations of pharmacologists and physiologists in respect to the influence of a change in the *milieu* on the oxidative capacity of cells are now receiving an interpretation more in accord with our newer conceptions and more capable of mathematical treatment. Thus, W. Mansfield Clark, who has distinguished himself in this field, points out in one of his recent papers how intimately related are the oxidation-reduction systems and acid-base systems of the animal organism, and adds:

> This has long been suspected to be a matter of profound importance in physiology, but it is believed that this (his) is the first systematic presentation among the numerous theoretical possibilities among the interrelated acid-base and oxidation-reduction equilibrium states.

I have also no time to comment on the reasons that induce the pharmacologist to follow your investigations with an absorbing interest and with keen enthusiasm. Some of these reasons are found in the biochemical problems outlined above, the essential parts of which are woven into the very texture of his thought, if I may say so. But quite aside from these more fundamental problems, there exists also a community of interests between us concerning the invaluable remedies with which chemists, working often in close cooperation with the pharmacologists, are enriching our *materia medica*. Occasionally, alas, in their cooperation in this field, both parties have fallen into the very human error of exaggerating the practical importance of their discoveries and researches—which latter, indeed, are often too solely and too transiently utilitarian in purpose and in character to redound to the credit of either party to the transaction.

In regard to the influence that our American predecessors and our own contemporaries in this country have had in shaping the future course of the biological and medical sciences I need not speak at length. Your influence, direct and indirect, on these sciences can be traced in the pages of the admirable historical review of your activities during the past half century published in the Golden Jubilee Number of *The Journal of the Ameri-*

can Chemical Society, though evidently the biochemical implications of this half-century's work could not always be brought out by the writers of that volume. As never before in our history, our younger biological and medical investigators are utilizing in their investigations the methods and principles of the various divisions of your science. A study of the various chemical, biochemical, biological, pharmacological, physiological and experimental medical journals of the country will furnish conclusive evidence that our countrymen are making fine progress in the broad fields covered by these journals.

PHYSICAL CHEMISTRY AND BIOLOGY

I come now to a consideration of some special points of contact between the physical chemist and the biologist, quite apart from the numerous references that have already been made to such contacts in earlier passages of this lecture. These contacts are conditioned by the composition and structure of the living substance—protoplasm—and the multifarious units—the cells of plants and animals—to which the protoplasmic structures are confined. The cell doctrine, far-reaching in its influence upon medicine and biology, as finally formulated by Dutrochet, Schwann and Schleiden before the middle of the last century, is to be counted among the very greatest discoveries of that century, comparable in its consequences for biology to those that followed the acceptance of the atomic and molecular hypothesis in chemistry.

The mysteries of life lie concealed in these units, individually of microscopic size only, and specialized in structure and function in the multicellular plants and animals. Naturally, I can not here comment on the reasons that necessitate a minimum external surface area for a cell, nor can I consider the influence of cells upon one another, nor the inadequacy of the cell theory to cover satisfactorily all the facts of development, as is claimed by a few biologists. The individual cell, wherever found, must be regarded as a congeries of minute, discrete chemical factories, standing in a possibly mobile spatial relationship to one another and working together in a beautifully harmonious manner. Surfaces, internal and external, and surface energies are of paramount importance in these individual units of life, as they are also when the units are aggregated in a manner to produce areas astonishingly large in dimension. The total internal surface of a given single cell, when the extensive colloidal interfaces, not to mention the limiting surfaces of the nuclei, nucleoli, chromosomes, plastids, zymogen granules, vacuoles or other microscopically determinable components, would, if computable, be found to be very large in comparison with that of its outer envelope. These internal surfaces, or boundaries between contiguous phases, are the seats of all manner of changes and operations, not merely all such as are broadly described as physico-chemical in character (electrical charges, etc.), but I venture to assert that all those protoplasmic processes that are usually defined as purely chemical in character, that is to say, oxidations, reductions, syntheses, hydrolytic decompositions, etc., are likewise surface actions. In the living cell, then, we have a system, the *microphysical* or *ultramicrophysical structure* of which differentiates it entirely from the systems of our test tubes and enables the cell to carry on all of its manifold processes at 37.5° C., more or less, as the case may be.

The labile oriented molecular arrangement in space and time of the surfaces of discontinuity of the phases of the various heterogeneous systems of the living cell and the constant interaction of these surfaces with sources of free energy of the environment, may account for the continued differentiations, apparently purposeful and directed, that characterize living things, as compared with the trend of non-living things toward less complex states. It may well be that an increase in our knowledge of molecular mechanisms in both fields will prove that the difference in behavior of living organisms and matter devoid of life is, in the last analysis, one of degree and not of kind.

I have said above that considerations of surface are of paramount importance in biology. Nature has found a means of producing very large surfaces by the aggregation of countless cells, as in leaves, root tendrils, capillaries and innumerable other structures. Krogh has calculated that if we suppose the total weight of a man's muscles to be 50 kilograms and his capillaries to number 2,000 per square millimeter of cross section, the total length of the capillaries of our muscles alone would be something like 100,000 kilometers, and their total surface 6,300 square meters. Our lungs, when collapsed, are small organs, but in them are found 725,000,000, more or less, of little pockets or alveoli where our thin-walled capillaries exchange carbon dioxide for oxygen. The total internal area of these lung terminals or alveoli is close to 100 square meters, enough for thirty suits of clothes, as Sir Arthur Shipley puts it in his valuable little book entitled "Life." The thin-walled capillaries of the lungs, one thousandth of a millimeter in wall thickness, make it possible for the slow process of diffusion to do the work necessary for the maintenance of life in less than the four seconds of each respiratory act. And so everywhere throughout the body we find these enormously extensive areas of cell surfaces—mechanisms for effective absorption, secretion and excretion in the unit of time.

I can not close this outline of "contacts" without a few further words on the bearings of physical chemistry on physiology and medicine. Every scientist of our day has learned that our countryman, J. Willard Gibbs, a man of the highest genius, by his fundamental and profound researches first placed the theory of surface phenomena on a truly scientific basis and made it capable of statistical and thermodynamical treatment. Since his day our knowledge of the actual molecular structure of the surfaces of both simple and heterogeneous systems has been greatly extended by scientists in this and in foreign countries—in this country by Langmuir, Harkins and others. Now, for more than half a century, physiologists and pharmacologists have been impressed, by the nature of the problems they were facing, with the need of more accurate knowledge of the physicochemical and chemical occurrences at the interfaces and limiting membranes of living structures. Naturally, their problems are bound up with questions involving adsorption and surface tension, colloidal sols and gels, the influence of the ever-varying chemical state of cell membranes, catalysis and related actions. Pharmacologists have always been confronted by the physico-chemical difficulties encountered in their attempts to analyze and understand the effects of drugs, including the powerfully acting hormones of our body, on both unicellular organisms and on the more complex structures of the animal body. My collaborators and I, to give but one example, have encountered such difficulties in our attempts to explain the increased susceptibility of the central nervous system of frogs to certain dyestuffs (ordinarily harmless) when administered subsequently to certain, otherwise trivial, injuries which so greatly increase the permeability of the capillaries and the adsorptive power of the cells of the brain and spinal cord that the ordinarily harmless dyestuff now reaches such a concentration in these cells (as can be shown by color tests) that violent strychnine-like convulsions immediately follow.

Workers in the medical sciences, as in biology, are bound to profit greatly, if not at present, then later, by the newer revelations in regard to the molecular structure of surfaces, more particularly those of micro-heterogeneous systems. They will agree with Professor Donnan when he gives expression to his conviction that "the newly recognized 'two-dimensional' molecular world . . . and the new knowledge of the structure of this 'surface world' presents phenomena of molecular orientation of the highest importance for the understanding of great regions of natural phenomena." Even though physical chemists are at present in disagreement in respect to many points in connection with this "two-dimensional" molecular world, the biologist must greet with enthusiasm every new and undoubted fact here discovered and the future theoretical developments in relation to it, even though he may only imperfectly comprehend the latter.

I realize fully that it is not within my competence to speak authoritatively in regard to the contacts between physical chemistry and vital occurrences, but I can not forbear citing, as a conclusion to my remarks on this subject, a heartening passage from Professor Donnan's address "On the influence of J. Willard Gibbs on the science of physical chemistry":

> In physiology the power and value of thermodynamical methods have been fully recognized only in comparatively recent times. Perhaps after another century of research in this science there may come another Willard Gibbs, who will discover the fundamental equations of the living cell, where

the unseen of the past seems to reach out and grip the future. But for that we shall require something more than linear differential equations.

The biologist, having in mind the ambitious attempts of the iatro-physicists and the iatro-mathematicians of the seventeenth century, as Borelli and others, to reduce physiology to physics and "to ornament and enrich it by mathematical demonstrations," and while not unmindful of the great value inherent in the contributions of these pioneers to haemodynamics and the mechanics of muscular movement, will feel, nevertheless, that the physical chemists of our day, like their mathematical predecessors three centuries ago, do not always take into account the complexities encountered in this laudable purpose of reducing life phenomena to fundamental equations. It is not specialized mechanisms, as the muscle, the selective permeability of membranes or certain definite chemical and physico-chemical processes like those of the respiration, which are already capable of thermodynamical treatment, that offer difficulties to the realization of this laudable ambition, but the more general cytological problems of biology such as are encountered in the study of cellular processes as a whole, of cell organization into highly differentiated structures, and of development in general, together with the difficult problems of heredity, that will tax the power of the Gibbses of the future. Or, consider the difficulties that will confront the mathematician of the future in his efforts to express the entire life processes and the reproductive powers of a single-celled organism only, as, say one of the paramoecia, in the form of valid general equations. To the biologist, it must appear that not one Gibbs but perhaps a half dozen or more will be required and their genius will be able to achieve results of value only after many more quantitatively determined biological facts shall be at their disposal than is now the case. The writer is an optimist in regard to the mental powers of the elect of our species, but he can not but feel that a single century will hardly suffice for the realization of these hopes, which biologists in general cherish equally with physicists and chemists.

ORGANS OF INTERNAL SECRETION

I may now be permitted to call your attention to a field of knowledge that has occupied experimenters for more than half a century—a field of great importance not only to the biologist in general, but to those who are associated in any way with medicine, as the bio-chemist, the physiologist, pharmacologist, the numerous representatives of the medical and surgical professions—a field that is concerned with a study of the functions, both normal and abnormal, of the organs of internal secretion. To the chemist is given a unique opportunity in this field and here, as is so often the case in biology, the last word is his. In saying this I have particularly in mind the obvious chemical aspects of the problems here presented, as, once they are cleared up, the physiologist and the physician are enabled to outline their own problems relating to the function of these organs with greater precision.

The significance of the interdependence of the various mechanisms of the animal body, of their admirably regulated activity and the harmonious manner in which these mechanisms cooperate in the development and growth of the individual from the moment when they first become apparent in early embryonic life to the time when we are returned to the dust from which we sprang has no doubt always been apparent to the mind of man, as is so well illustrated in Aesop's fable that it will not do for the various members of the body to fall out with each other. The medicine of an older time has long used the words *consensus partium*, the mutual dependence on each other, in other words, this interrelationship, of the various organs. Generally speaking, until about a century ago the *consensus partium* was supposed to be effected solely through the intermediation of the nervous system—a point of view tersely expressed in 1752 by the French anatomist Cuvier when he said: "Le système nerveux est, au fond, tout l'animal, les autres systèmes ne sont lè que pour le servir." According to this view, then, to put the matter in popular language, certain control stations in the brain and spinal cord and of the peripheral autonomic nervous system coordinate all the functional activities solely by means of telegraphic communication (though a nervous impulse is quite a different thing from an electric current). During the past seventy years, however, conjoined chemical and physiological discoveries have brought to light a new method of communication between organs and a coordination of their functional activities, closely linked, to be sure, with the longer known type of control but yet quite distinctly different. To-day we also have, as Professor Starling so aptly put it, an efficient postal service at the disposal of the various mechanisms of our body. The little packets of chemicals sent out by the organs of internal secretion are carried by the blood stream to their appointed destinations and are generally called hormones, the name given to them by Starling twenty years ago. The term is derived from the Greek verb ὁρμάω (I stir up or excite), and is perhaps less appropriate than the designation *chemical messenger*, earlier applied to this class of substances by the same physiologist.

These chemical messengers may initiate changes in the functional state of a distant nerve cell or nerve terminal, or a series of chemical changes, or take part in some intermediary chemical reactions in a distant organ—reactions that may or may not have been initiated primarily by a preliminary nervous impulse or telegraphic message.

The organs in which these chemical messengers are elaborated and from which they are despatched into the blood stream are known as organs of internal secretion, or endocrine organs. Some of these are called ductless glands—structures that are entirely devoid of secretory ducts, as their name implies, and can only perform their functions by sending their chemical messengers into the blood and lymph, by which they are transmitted to the various organs of the body. Examples of such ductless glands are the thyroid, the parathyroids, the pituitary organ and the intestinal mucosa. Examples of endocrine organs that play the double rôle of producing ferments or other chemical agents that are eliminated by special ducts or passages as well as chemical messengers that pass into the blood stream are the pancreas and the sex glands.

The entire list of organs possessing an endocrine function need not be given here. Let it suffice to say that additions to the list of chemical messengers are constantly being made as the conceptions in this department of physiology have broadened in consequence of more intensive experimentation. Organs such as the heart, for example, have been shown to produce substances that influence the rate of its contractions by an action on its regulartory nervous mechanism. The carbon dioxide that is excreted by our tissues as an end product of their metabolic activity acts as a hormone or regulator of the external respiratory apparatus. In the widest and perhaps truest sense of the word, every tissue, as Schäfer remarks, is an internally secreting structure.

Juice expressed from the embryonic heart of the chick contains a chemical substance, or perhaps more than one, that is indispensable for the continued growth, in the warm chamber, of an isolated fragment of an embryonic heart. This continued maintenance of life and growth in the thermostat of fragments of tissues excised from cold or warm blooded animals or from tumors was initiated by Ross G. Harrison and is known as tissue culture. Carrel has suggested the term "Trephones" (from τρέφω—I feed) for the growth-promoting substances of embryonic tissue juice and of leucocytic extracts. I have called to your mind the work of Carrel and his associates on the artificial propagation of the fibroblasts taken from a fragment of the embryonic heart of the chick because the indispensable trephones or growth-producing substances that must constantly be added to the culture medium may properly be called hormones. They are produced in the cell laboratories of the chick's heart and function in some as yet unexplained manner as regulators of the complex intermediate chemical processes that are necessary to the orderly life of the cell. It will interest you in this connection to learn that Carrel and his collaborators have, from January, 1912, to June, 1927, carried a fragment of embryonic heart tissue through 2,987 generations or "passages of cultivation." There is no reason to doubt that this fragment of tissue would continue to develop indefinitely, provided only the temperature of the thermostat be maintained at 37.5° and that the composition of the nutritive medium remains constant. These extraordinary growth-promoting substances or catalysts of the embryonic tissue (and of leucocytes) may for the present be classed with the hormones.

You are all aware that modern research has revealed the astonishing fact that we require for the complete nourishment of the body, in addition to the energy-yielding and mineral constituents of our food, minute quantities of other substances which are called vitamins. There are many analogies between the physiological properties of these vitamins, which may be called hormones of plant origin, and those that are produced in the animal organism.

According to many plant physiologists, chemical messengers appear to play an important, if not occasionally a greater, rôle in the life of plants than they do in that of animals, but as this phase of the subject lies outside the field of my own experimentation I shall content myself with having called it to your attention.

I can not enter into the details of the story of the discovery of the functions of the ductless glands proper or of the endocrine functions of the organs that have both an external and an internal secretion. The whole subject constitutes one of the greatest contributions of the nineteenth century to scientific medicine. Osler in his "Evolution of Modern Medicine" gives it as his opinion that

> there is perhaps no more fascinating story in the history of science than that of the discovery of the so-called ductless glands. . . . No such miracles have ever been wrought by physicians as those which we see in connection with the internal secretion of the thyroid gland. The myth of bringing the dead back to life has been associated with the names of many great healers since the incident of Empedocles and Pantheia, but nowadays the dead in mind and the deformed in body may be restored by the touch of the magic wand of science. The study of the interaction of these internal secretions, their influence upon development, upon mental processes and upon disorders of metabolism is likely to prove in the future of a benefit scarcely less remarkable than that which we have traced in the infectious diseases.

We are but at the beginnings of knowledge, especially as concerns the chemical and physiological problems that are presented in this great field. Consider, for example, that remarkable and still unknown hormone elaborated in the anterior lobe of the hypophysis and passing from thence into the blood. In it we have a chemical agent that can affect the growth of bone and other structures to an extraordinary degree. The unchecked action of this growth-stimulating principle, extending over a period of years, leads to acromegaly or gigantism. Evans and Long, by injecting potent extracts of the anterior lobe of the hypophysis daily into young rats, have succeeded in producing veritable giants of their species. One typical experiment may be cited: a rat received intraperitoneally anterior hypophyseal substance for a period of 333 days. At the end of that time the animal weighed 596 grams, while its healthy litter mate control weighed only 248 grams.

Studies of this nature have succeeded for the first time in throwing a bridge across the chasm that has hitherto separated bio-chemistry from morphology. Distinguished anatomists, indeed, as Sir Arthur Keith and Professor Bulk, of Amsterdam, have expressed their conviction that the differentiation of mankind into racial types is due to the differential interaction of these endocrine organs. The cumulative experience of a host of medical observers during the past seventy-five years has demonstrated beyond a doubt that innumerable departures from the normal in respect to bodily stature, facial configuration, sexuality, general metabolism and even the mentality find their explanation in the over- or underactivity of these, anatomically speaking often quite insignificant, structures, or in a lack of harmony in their cooperation. It is one of the tragedies of life, a decree of fate, that we should be both "the beneficiaries and the victims of the chemical activities and correlations of our endocrine organs."

Our chemical knowledge of the elusive principles, elaborated in minute quantities only by these indispensable organs, is in its infancy, as I have already intimated, and still lags far behind our acquaintance with their physiological actions. Only two of them, substances of low molecular weight and relatively simple structure, have been prepared synthetically. In respect to these two, then, the organic chemist has again come to our rescue and both the experimentalist and the manufacturer are now free to accept their deliverance from the products of the slaughter house.

The first of these hormones to be conquered is the one that is elaborated in the so-called medulla of the suprarenal capsules, small yellowish structures, each shaped like a cocked hat and fitting snugly on top of its corresponding kidney. The two organs together, in a fully developed man, weigh less than a third of an ounce. Life is impossible without them. Like some others of the endocrine organs, notably the hypophysis, they are double structures, fused in the higher animals into what is apparently only one organ consisting of a medullary or inner portion and a cortical or outer portion. This cortical portion contains a hormone, or hormones, more immediately necessary to life than that produced in the medulla. Investigators are now occupying themselves with it and we look forward, I confidently believe, to a successful outcome of their researches.

The medullary hormone is called by various names, as adrenin, suprarenalin, suprarenin, adrenalin and epinephrine, the latter having been adopted by the United States Pharmacopoeia as the official designation. This name was coined by me thirty years ago at a time when I supposed that the form in which I had succeeded in isolating it represented the base as it actually exists in the capsules.

Without going into the details of earlier attempts to isolate the principle, all of which I have described in a historical paper published in 1903 (*Amer. Jour. Pharm.* 1903, *75*, p. 301), I hope that you will permit me to say a few words about my own investigations towards its isolation. With the assistance of the late A. C. Crawford I succeeded in separating the hormone from its numerous tissue concomitants in the form of a benzoyl derivative. On decomposing this benzoyl derivative with hot dilute sulphuric acid in an autoclave we obtained the active principle in the form of a sulphate which possessed the characteristic physiological activities of suprarenal extracts and reacted, furthermore, with a series of chemical reagents in a manner that is quite specific for such extracts and limited to them. The principle as obtained by saponification of the benzoyl derivative was thrown out of its solution by means of ammonia in the amorphous state and was shown to be a weak base. A picrate, a bisulphate and other salts of it were prepared, all of which were shown to possess a high degree of physiological activity. An acetyl derivative, a phenylcarbamic ester and other derivatives were also prepared and certain degradation products of the base were isolated and studied. Without giving any further details of this earlier work, which occupied my time for a number of years, I will merely state that the elementary composition of the base was established by analysis of several derivatives, including a sulphate, and was stated to be represented by the formula $C_{17}H_{15}NO_4$ (*Zeit. f. physiol. Chem.*, 1899, xxviii, 318).

After I had completed the above described investigations and while I was still endeavoring to improve my processes I was visited one day in the fall of 1900 (as I recall it) by the Japanese chemist, J. Takamine, who examined with great interest the various compounds and salts of epinephrine that were placed before him. He inquired particularly whether I did not think it possible that my salts of epinephrine could be prepared by a simpler process than mine, more especially without the troublesome and in this case wasteful process of benzoylating extracts of an animal tissue. He remarked in this connection that he loved to plant a seed and see it grow in the technical field. I told Takamine that I was quite of his opinion that the process could no doubt be improved and simplified. At this very time also, v. Fürth had just prepared an amorphous, highly active, indigo-colored compound of the active principle, which he named suprarenin, but no analytical data were given and no empirical formula for his principle was established.

Takamine prepared suprarenal extracts more concentrated than mine, and without first attempting to separate the hormone from its numerous concomitants by benzoylating or otherwise, simply added ammonia—the reagent that I had so long employed—to his concentrated extracts, whereupon he immediately obtained the native base in the form of burr-like clusters of minute prisms in place of my amorphous base. I have often been asked why I had not myself attempted to solve the problem in this very simple fashion. The truth is that I had tried to do so but always found that the dilute extracts tested simply turned pink in a short time on the addition of ammonia without depositing a base, either crystalline or amorphous. Inasmuch as even very dilute solutions of the salts obtained by me on saponifying the benzoyl derivatives always gave a precipitate with ammonia, I fell back on the hypothesis that other constituents of the impure extracts prevented its precipitation by ammonia from my dilute native extracts—an erroneous assumption. Takamine's success was due to the employment of ammonia on very highly concentrated, though impure, extracts. The fact that my amorphous base could be precipitated from even highly dilute solutions was, as I soon found, due to the fact that one benzoyl radical had not been removed during the saponification. Takamine adopted the empirical formula $C_{10}H_{15}NO_3$ as the "probable empirical formula" of his substance, which was immediately patented in this country and manufactured, greatly to the advantage of medicine. I was soon able to demonstrate that my epinephrine—$C_{17}H_{15}NO_4$—had retained a single benzoyl radical, C_6H_5CO, that had resisted saponification and could only be removed from the base by drastic treatment with strong acids and heat, a treatment which at the same time obliterated every trace of the characteristic physiological action of the hormone (Johns Hopkins Hospital Bulletin, 1901, vol. xii, p. 337). I suspect that the retained radical was attached to the imide nitrogen of the side chain of the molecule—an unusual circumstance in any event. Subtracting the molecular weight of the retained radical from my original formula, $C_{17}H_{15}NO_4$, leaves $C_{10}H_{10}NO_3$, which formula is very close indeed to that assigned by Takamine to his crystalline base, and Aldrich, who had been my assistant and who, coincidently with Takamine and quite independently of him, also discovered that the base is obtainable in crystalline form when ammonia is added in sufficient quantity to a highly concentrated suprarenal extract, wrote, without knowing at the time that I had already discovered the concealed radical: "It is interesting to note in this connection that if we subtract a benzoyl residue from Abel's formula for epinephrin—$C_{17}H_{15}NO_4$—we obtain a formula $C_{10}H_{10}NO_3$ which is not far removed from that of adrenalin." (*Amer. Journ. Physiol.*, vol. V, p. 461).

I venture to say in extenuation of the blunders of a pioneer in this field that the results obtained by me and my close approximation to the true elementary composition of the hormone were not due to chance, but could have been obtained only from the study and analysis of a series of fairly pure chemical individuals. Every one of these derivatives had, however, as already stated, retained a single benzoyl radical. The efforts of years on my part in this once mysterious field of suprarenal medullary bio-chemistry, marred by blunders as they were, eventuated, then, in the isolation of the hormone, not in the form of the free base but in that of its monobenzoyl derivative. Aldrich finally established the true empirical formula, $C_9H_{13}NO_3$, the correctness of which was afterwards conclusively verified by others abroad.

I can not look back on my own poor efforts to elucidate the chemical constitution of the compound with pleasure. After the preparatory pioneer work thus far outlined there followed the brilliant researches, in respect to the chemical constitution of the hormone, of the chemists, Dakin, Jowett, Pauly, Friedmann, Stolz and Flächer, which have finally culminated in the synthetic production, first of the racemic and later of the laevo-rotatory form, as produced in the animal organism itself. The work of these chemists has shown that the suprarenal medullary hormone is an aromatic amino-alcohol, dihydroxymethyl-amino-ethylol-benzene,

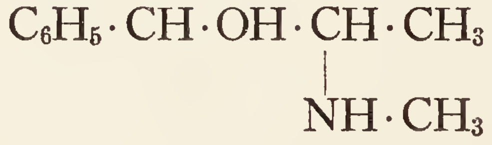
$$C_6H_3(OH)_2 \cdot CHOH \cdot CH_2 \cdot NH \cdot CH_3.$$

The cells of the medullary portion of the suprarenal gland are intimately related in their origin to the sympathetic nervous system, and we are not surprised therefore to find their secretory product, adrenalin or epinephrine (U. S. P.), has a pharmacological and quite specific affinity for the sympathetic nervous system—the thoracic-abdominal part of the autonomic nervous apparatus. The changes induced by epinephrine in the activity of the various organs innervated by this system are in all respects like those that are brought about when the sympathetic fibers controlling these organs are stimulated by an electric current. A small quantity of the hormone administered intravenously will cause vaso-constriction, increased rapidity of heart action, dilatation of the pupil, relaxation of constricted bronchioles, inhibition of the peristaltic movements in the alimentary canal, contraction of the pyloric and ileo-coecal sphincters, increased motility of the pregnant uterus and mobilization of the glycogen of the liver with a resultant glycosuria. We have here then an example of a definite chemical principle of known structure which, when carried by the blood to a terminal point of the sympathetic system, induces exactly those alterations that follow electrical stimulation of the sympathetic fibers that pass to these terminal points. In more technical pharmacological terms, the hormone stimulates, sensitizes or acts in an inhibitory manner on sympathetic myoneural or adenoneural, junctions of the sympathetic nervous system. Because of this highly specific action this hormone and the many others of its class that have been isolated in recent years from various animal, plant and bacterial products have been given the name of sympathomimetic amines.

I can not pause to give an account of the various beneficial uses which several of these sympathomimetic amines have found in medical practice. Nor can I enter upon a chemical or pharmacological analysis of these numerous, naturally occurring or synthetically prepared, biologically very important substances that are classified with epinephrine as phenylalkyl- or phenylalkanolamines. This group is only one of many groups of physiologically more or less active substances grouped together as a class of biogenous amines.

It is worth while pointing out in this connection that from the structural point of view a whole series of alkaloids, as hydrocotarnine, anhalamine, anhalonidine, papaverine, laudanine, bulbocapnine, corydine, berberine, canadine, cryptopine, protopine and many others can be brought into genetic relationship with the phenylalkylamines above referred to, as has been outlined by Elger.

I may conclude this section of my lecture by a brief description of a plant principle, ephedrine, closely related in structure and physiological action to the hormone of the suprarenal medulla. For more than 5,000 years the Chinese have used the stems of *Ephedra vulgaris*, under the name of Ma huang, as a medicine famed among them as a diaphoretic, a circulatory stimulant, a sedative in cough and an antipyretic. The Japanese chemists Yamanashi and Nagai first isolated from Ma huang a derivative of phenylethylamine and named it ephedrine. The chemical structure of this laevo-gyrous plant base has been determined and is evident from its formula,



$$C_6H_5\cdot CH\cdot OH\cdot \underset{\displaystyle NH\cdot CH_3}{\underset{|}{CH}}\cdot CH_3$$

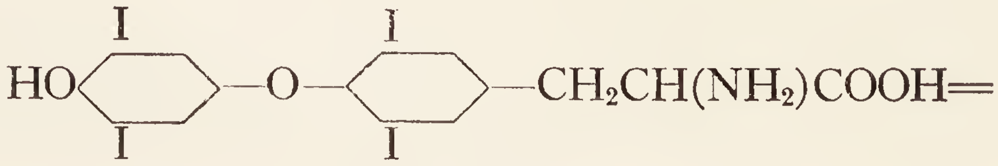
1-phenyl-1-hydroxy-2-methylaminopropane. It will be noted, on comparing this structural formula with that of epinephrine, that ephedrine contains two asymmetric carbon atoms as compared with the one of epinephrine, which latter also differs in containing two phenolic hydroxyl groups in the ortho-position to each other, making it much more susceptible to oxidation than ephedrine. Ladenburg and Oelschlägel, who determined the chemical structure of ephedrine, have succeeded in isolating from the *Ephedra vulgaris* of Europe a stereoisomer, pseudo-ephedrine, which is not, strictly speaking, an optical antimere in its configuration. Its dextrorotation is due to a difference in the spatial arrangement of the alcoholic hydroxyl group of the above structural formula. The four possible isomers of ephedrine have been prepared synthetically and isolated and an extensive literature has already appeared in connection with their chemical and pharmacological properties and with the medical uses of the laevo-gyrous isomer, ephedrine. The pharmacological action of this plant phenylalkanolamine is qualitatively identical with that of the suprarenal base, epinephrine; in other words, it is a typically acting sympathomimetic amine. It is less active physiologically than the latter, but its effects are more prolonged. In the form of ephedrine sulphate it has already been found to be of great value in nasal operations and in ophthalmology. Its ultimate value in serious diseases of the heart, in shock, hypotension and other pathological states is now being made the subject of numerous investigations.

THE HORMONE OF THE THYROID GLAND

A second hormone, the chemical constitution of which has only recently been unravelled, is that produced by the thyroid gland, a small trilobed organ set astride the windpipe, below the larynx, and weighing in man 35 grams or 1¼ ounces. In this connection I may add that we are also endowed with a number of very small endocrine organs, the parathyroids, usually four in number, two on each side of the neck, closely adherent to the dorsal surface of each lateral lobe of the thyroid gland.

While the thyroid gland of man weighs about 35 grams or $1\frac{1}{4}$ ounces, the four parathyroids, each about the size of a hempseed, weigh together only half a gram or 7 to 8 grains. These minute glands, like the thyroid itself and most of the other organs of internal secretion, have an unusually good blood supply and are essential to life, one of their known functions being that of controlling in some as yet undefined manner the chemical combinations of the calcium in the tissues. Their hormone has not yet been isolated.

The hormone of the thyroid gland was first isolated in crystalline form by E. C. Kendall in 1914, and was named thyroxine by him. A year ago C. H. Harington, of University College Hospital, London, described an improved method for the separation of thyroxine from the thyroid tissue and by the brilliant application of well-known methods of break-down and synthesis obtained an iodine-free degradation product, desiodothyroxine, differing only from the natural hormone in being devoid of the four iodine atoms present in the latter. Harington next succeeded in working out the constitution of this desiodothyroxine and found it to be the p-hydroxyphenyl ether of tyrosine. Next, Professor Barger and he conjointly prepared in a masterly manner a series of organic derivatives which were utilized in finally effecting the synthesis of the hormone in its racemic form. The brilliant work of these investigators, carried out along classical chemical lines, has now incontestably established the constitution of the hormone as being β-[3,5-diiodo-4-(3′,5′-4′-hydroxyphenoxy) phenyl]-α-aminopropionic acid, as represented by the following formula:



$$HO\overset{I}{\underset{I}{\bigcirc}}{-}O{-}\overset{I}{\underset{I}{\bigcirc}}{-}CH_2CH(NH_2)COOH=$$

$C_{15}H_{11}O_4NI_4$

Synthetic thyroxine, as prepared by Harington and Barger, is obtained in the form of a crystalline precipitate consisting of rosettes and sheaves of colorless needles. The compound is insoluble in water and the usual organic solvents, soluble at room temperature in solutions of the alkali hydroxides, provided their concentration be not too high. It dissolves in sodium carbonate solution only on boiling and is soluble in 90 per cent. alcohol containing either an alkali hydroxide or a mineral acid. Sodium, ammonium and barium salts have been prepared which agree in their properties with the corresponding salts of the natural product. The identity of the synthetic product with the natural hormone has thus been conclusively established by the brilliant researches of Harington and Barger.

Professor D. Murray Lyon, of Edinburgh, has studied the effects of synthetic thyroxine on two myxoedematous patients whose basal metabolism rates were respectively 32 and 45 per cent. below normal. After intravenous administration to each of the two of a total dose of 14 mgs. of the hormone, given in divided doses over a period of six days, the basal metabolism of the one rose to within 6 per cent. of normal and that of the other to 3 per cent. above normal. It is concluded, therefore, that the effect of synthetic thyroxine in raising the basal metabolic rate of these two patients is quantitatively similar to that reported by Boothby and Sandiford, of the Mayo Clinic, in 1924, for natural thyroxine.

THE PANCREAS AND ITS HORMONE, INSULIN

Of the endocrine glands which are now being so actively investigated, I have selected for further discussion the pancreas and its internal secretory product, insulin. Needless to say that within the brief time left I can do no more than just touch on a few of the important researches that constitute the historical background for our present conception of the indispensable rôle of this gland in carbohydrate metabolism. There are four important milestones in the history of carbohydrate metabolism which can receive but the barest mention here.

I. The epoch-making researches of Claude Bernard in relation to the functions of the liver and pancreas and more especially with reference to their rôle in carbohydrate metabolism constitute the first of the four milestones. Bernard, in his masterful and logical way, worked out the fact that the liver is capable of polymerizing dextrose into a starch—or dextrin-like substance which he named glycogen because the hepatic tissue is able, through the agency of a ferment, to reconvert it into sugar. His findings caused Bernard to view the liver as an organ of internal secretion (and he was one of the first to use this term), that is, an organ which manufactures in its cells a product which can be converted into a substance transportable by the blood to all parts of the body and utilizable by the tissues. Bernard, not content merely with proving the existence of such a product in the liver, set out to extract and to isolate it from this organ in pure form, a feat which he accomplished in 1857 after many years of preliminary work. Having now isolated glycogen, Bernard determined the conditions under which it is formed and is reconverted into sugar, and before long he was able to show that the reciprocal relationship between glycogen and glucose in the animal organism is entirely analogous to that existing between starch and sugar in the metabolism of plants. The name animal starch, by which glycogen is frequently called, is therefore well taken.

II. The second important development in this field came when the pathologist Langerhans, then a young investigator in Virchow's laboratory in Berlin, discovered (1867–69) that there are contained in the pancreas groups of cells situated between the acini and markedly different from those of the ordinary glandular type. These groups, usually round, are composed of small, irregular, polygonal cells with a round nucleus and a homogeneous refractive cell body. Numerous observers have verified this discovery and they have been designated ever since as the islands of Langerhans or the islet tissue. These islands of Langerhans are the seat of formation of the pancreatic hormone, which has been appropriately named insulin in accordance with the suggestion first made by the Belgian, de Meyer, in 1909 and independently by the distinguished physiologist, Schäfer, in 1916 and adopted at the latter's suggestion by the Toronto workers. But the proof that the islet tissue of the pancreas produces the hormone was not established until near the close of the last century, and it required the joint labors of many investigators to establish this point with certainty.

In certain teleostean fishes, the islets, homologous in structure and in their function with those of Langerhans, exist as organs quite separate from the pancreas, and an additional link in the chain of evidence in support of the theory that the islet cells alone can produce insulin was furnished by the Toronto investigators, Macleod and his associates, who extracted the hormone from these specialized teleostean organs and found that it possessed the physiological properties of the insulin prepared from the pancreas of beeves. The islets are more abundant in the pancreas of the higher animals than was formerly believed to be the case. In the entire pancreas of the guinea pig, according to Bensley, as many as 56,000 islets have been counted, "so that the endocrine tissue, instead of being rather scant, as it has usually been thought to be, is rather abundant" (Macleod).

III. A third great step forward was taken in 1889–91. In those years the clinicians v. Mering and Minkowski discovered that complete removal of the pancreas from dogs is followed by a diseased state which is practically in all respects like that seen in human diabetes mellitus. This clean-cut piece of experimental work, with the consequences that are logically deducible from it, constitutes one of the greatest achievements in this field since the discovery of glycogen and the demonstration of its physiological properties by Claude Bernard fifty years before. These consequences, indeed, played the chief rôle in the establishment of the proof outlined above under the heading (II) that the islets of Langerhans are the seat of formation of insulin, and furthermore, when taken in conjunction with the subsequent findings of pathologists, as Opie and others, they forced on men's minds the conviction that the cause of that frequently occurring and serious disease, diabetes mellitus, must be referred to an inadequate functioning of the islets.

The crucial demonstration by v. Mering and Minkowski that ablation of the pancreas leads inevitably to diabetes naturally served as a great impetus to further research which confirmed their work and paved the way for the fourth great historical event in this field, presently to be described. Leaving out here all reference to the investigations of about twenty workers in the decade or two following the discovery of v. Mering and Minkowski, let me speak very briefly of the results obtained by workers in this field closer to our own time, results that almost succeeded in giving us insulin 15 years ago. The Toronto investigators have, in a very generous spirit, given full credit to the unsuccessful attempts of their predecessors to prepare serviceable extracts of the pancreas. In 1908 Zuelzer prepared an alcoholic extract from the pancreas of recently fed animals and obtained rather striking results from its use on a pancreatectomized dog and on eight diabetic patients. It is interesting to note, in studying Zuelzer's protocols, how greatly both the output of urinary sugar and of the acetone bodies was reduced in his patients as the result of the intravenous injection of his preparation. Untoward symptoms, such as a rise of temperature and chills, appeared both in Zuelzer's patients and in those of Forschbach, another clinician who tried Zuelzer's extract. In the opinion of the latter, the alleviation of the symptoms in diabetic patients was due rather to the febrile reaction induced by the injections than to a specific action of the hormone assumed by Zuelzer to be present in his extracts, and so it fell out that the successful use of pancreatic extracts for the treatment of diabetes has had to wait until our day for men equipped with newer methods and who, above all else, based their conclusions both on evidence obtained from well controlled animal experimentation and on clinical experience with diabetic patients.

E. L. Scott, who worked in this city in 1911–12 in Professor Carlson's laboratory, also came very close to obtaining a pancreatic extract that might have been serviceable, had it been tried out on human beings, since, by his method of preparation, blood-pressure lowering substances were removed. Scott's extracts, when injected intravenously into completely pancreatectomized dogs, diminished temporarily the sugar excretion and lowered the D:N ratio of the urine. Unfortunately, however, this investigator did not properly interpret his findings, inasmuch as he concluded that it does not follow that the effects induced by his extracts were due to the presence in them of the internal secretion of the pancreas. In 1913 Murlin and Kramer, in their study of the effects of pancreatic extracts on glycosuria, were led to the conclusion that neither their extracts nor the transfusion of normal blood "is as yet of any practical importance in restoring to the depancreatized dog the ability to burn sugar." Kleiner, using emulsions of the dog's pancreas infused slowly into a vein of a depancreatized dog, observed a temporary but marked decrease in the blood and urinary sugars and regarded his findings as furnishing evidence in support of the endocrine theory of experimental diabetes.

Other attempts to prepare an extract of the pancreas that would be serviceable in lowering the blood and urinary sugars and in ameliorating the symptoms of depancreatized animals and of human diabetics can not be detailed here.

IV. The numerous discoveries since Claude Bernard's day, grouped together under three periods as above, constituted the indispensable foundation for a fourth step—the preparation of an effective and stable extract that would unfailingly, or with rare exceptions, restore completely to health persons, old and young, sufferers from and often the early victims of that hitherto unconquerable malady, diabetes melitus.

All the world knows of the brilliant achievements of Banting, Best, Macleod and Collip and their collaborators, acting on an original suggestion of Banting, that have led to the fourth great epoch in the combined fields of bio-chemical, physiological, pharmacological and clinical investigation. The results of these talented investigators have been and will continue to be of incalculable value to mankind, and have opened up many new possibilities for the study and better comprehension of the difficult field of carbohydrate metabolism.

This brings me now to a brief account of my own endeavors and those of my collaborators to isolate and separate the true insulin hormone from its numerous concomitants in the therapeutic preparations now employed, serviceable as they are, in the treatment of diabetics. It is a proper aim of the scientist, a mandate even, if I may say so, is laid upon him, wherever it is humanly possible, to isolate and to identify the elusive and indispensable hormones from their complicated mixtures (messes, the chemist would say) in which nature presents them. Once this aim has been realized and the hormone has been separated as a well defined chemical individual, the next steps, such as the study of the constitution of the hormone and its eventual synthesis, if ultimately possible, fall to the chemist. As in all previous instances, the isolation of a hormone as a chemical individual gives to the biochemist and physiologist a cleaner approach for the solution of their problems than when they are compelled to use mixtures of unknown composition.

These studies were made possible through a generous grant from the Carnegie Corporation of New York. The earlier ones are set forth in the following publications from my laboratory: Abel and Geiling, *Journ. Pharmacol. and Exper. Therap.*, 1925, xxv, 423; Abel, Geiling, Alles and Raymond, SCIENCE, 1925, xlvii, 169. They deal chiefly with establishing the fact that sulphur in a labile form is an integral part of the insulin molecule and that the physiological activity goes hand in hand with the labile sulphur content of the molecule. I next succeeded in obtaining insulin in crystalline form and during the past year its preparation has been so simplified that, starting with commercial preparations such as the dry powder manufactured by the Connaught Laboratories of Toronto (13 units per milligram) or the concentrated liquid extracts furnished by Eli Lilly and Co. and E. H. Squibb and Sons (250–450 units per cc.), the crystals can be obtained in any desired quantity within a very few days. The salient features of the new method, the full details of which are given in *The Journal of Pharmacology and Experimental Therapeutics*, 1927, xxxi, 65, can be seen from the following brief description of a typical experiment in which, starting with 2.001 grams of a Toronto powder evaluated at 13 units per milligram, there was obtained a total of 0.5284 gram of crystalline insulin. Various samples of these crystals have been submitted to the insulin committee at Toronto for standardization, but owing to the press of other work the committee has not as yet been able to make a report on them. Our own standardizations of a recrystallized preparation against the International Standard Powder gave us a conservative value of 40 international units per milligram.

To the powder, dissolved in 20 cc. of 10 per cent. acetic acid, was added 80 cc. of brucine acetate solution (1 gram of base in 18 cc. of N/6 acetic acid) and then 40 cc. of 13.5 per cent aqueous pyridine; the resulting "pyridine precipitate" was centrifuged off and the clear fluid treated with 40 cc. of 0.65 per cent aqueous ammonia. The "ammonia precipitate" so obtained was likewise centrifuged off and the fluid set aside to crystallize in an Erlenmeyer flask. Next morning the walls and bottom of the flask were found lined with crystals which, after washing and drying, weighed 0.2776 gram. The "pyridine" and "ammonia" precipitates treated in the same way gave further crops of 0.1458 and 0.0614 gram, respectively, and a final crop of 0.0436 gram was obtained from the residue left by evaporating in a current of air at room temperature the mother liquors from the preceding fractions. The total yield of crystals was thus 0.5284 gram.

Starting with liquid preparations, the procedure is the same except that the crude insulin is first precipitated with insulin as described in earlier papers.

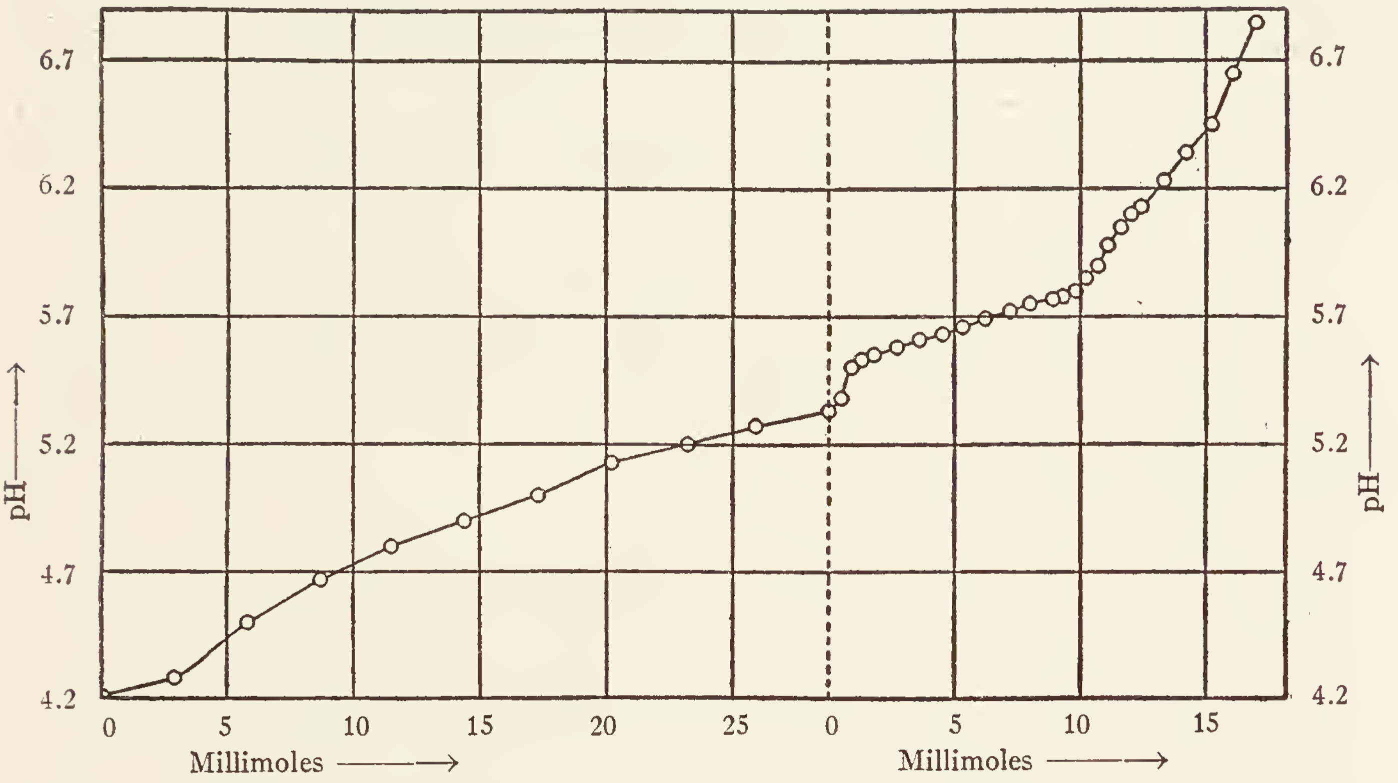


Fig. 1. Titration Curve showing pH Values of a Mixture of 40 cc. of Brucine Acetate Solution (1 Gram of Brucine Alkaloid Dissolved in Each 18 cc. of 0.132 n Acetic Acid) and 10 cc. of 1.317 n Acetic Acid upon the Addition of Pyridine amd Ammonia.

A total of 20 cc. of 1.441 n pyridine was added (left portion of curve), followed by 38 cc. of 0.444 n ammonium hydroxide (right side of curve). The amounts of the bases added are plotted in millimoles. The hydrogen-ion concentrations were estimated colorimetrically with Michaelis' nitrophenol indicators, using 0.1 cc. of buffer mixture for each determination.

Measurements with the Michaelis' nitrophenol indicators as well as with the quinhydrone electrode showed that the pH of the solution from which the insulin separated in crystalline form was 5.55–5.65. After centrifuging off the "ammonia precipitate" it may be necessary to add a little more ammonia to the fluid to bring it to the proper hydrogen-ion concentration before setting it aside to crystallize. The accompanying curve shows how the pH of a mixture of acetic acid and brucine, made up in the proportions employed in this method, varies with the gradual addition of the usual amounts of pyridine and ammonia.

The crystals are apparently dimorphous and fall into two general groups: (1) Crystals with well-defined double refraction, of negative character, with several habits, in the rhombohedral class; (2) crystals of a more equant habit, often with clearly defined crystal edges and no double refraction.

They give the Pauly, Millon, biuret and ninhydrin reactions but not the Voisonet, Hopkins-Cole or Acree tests for tryptophan or the Sullivan test for free cystine and cysteine.

The many solutions (in acetic acid, hydrochloric acid and ammonia) examined polarimetrically were always found to be laevo-rotatory, the magnitude of the rotation varying widely with the concentration and pH of the solution and with the nature of the solvent. For example, one preparation in hydrochloric acid showed a specific rotation of $-40°$; another, twice recrystallized, gave $-30°$ in N/6 acetic acid and $-17°$ in 0.011 N hydrochloric acid; with another in 0.65 per cent. ammonia the rotation was $-48°$ and changed in the course of several days through a maximum at $-63°$.

Numerous microanalyses on various preparations gave very concordant results agreeing closely with the empirical formula $C_{45}H_{69}O_{14}N_{11}S$ in the case of material dried at 105–20° in nitrogen under low pressure and $C_{45}H_{75}O_{17}N_{11}S$ (or $C_{45}H_{69}O_{14}N_{11}S \cdot 3H_2O$) for air-dried preparations; the labile or so-called "carbonate" sulphur content of the latter is about 1.10 per cent. or approximately 37.5 per cent. of the total sulphur. No satisfactory solvent for molecular weight determinations has yet been found.

No evidence has ever been obtained which would indicate that the crystals are not a homogeneous substance crystallizing in different types but a mixture of two substances, only one of which is physiologically active but both having the same solubilities and identical or nearly identical empirical compositions.

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